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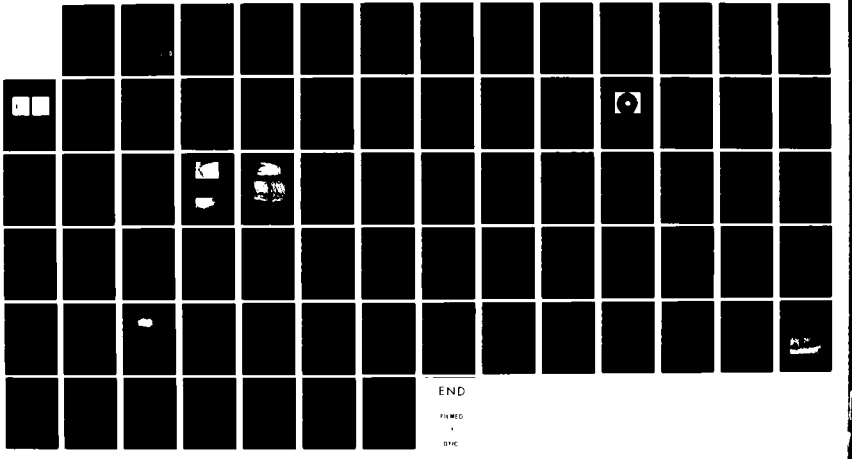

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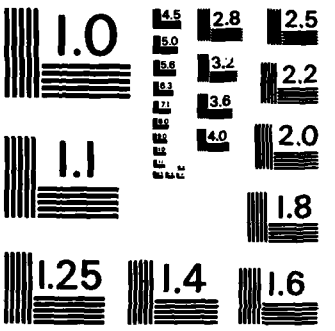

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COMBAT OCULAR PROBLEMS

Supplement *April 1982*

EDWIN S. BEATRICE, M.D.
Colonel, Medical Corps, United States Army
Editor-in-Chief

Sponsored by
DIVISION OF OCULAR HAZARDS
LETTERMAN ARMY INSTITUTE OF RESEARCH
COL John D. Marshall, Jr., MS
Commanding

Supplement to PROCEEDINGS OF CONFERENCE conducted October 20-21, 1980

LETTERMAN ARMY INSTITUTE OF RESEARCH
PRESIDIO OF SAN FRANCISCO, CALIFORNIA 94129

COMBAT OCULAR PROBLEMS
Supplement to *Proceedings of Conference* conducted October 20-21, 1980
April 1982

In conducting the research described in this report, the investigation adhered to the "Guide for the Care and Use of Laboratory Animals," as promulgated by the Committee on Revision of the Guide for Laboratory Animal Facilities and Care, Institute of Laboratory Animal Resources, National Research Council.

Investigators adhered to AR 70-25 and USAMRDC Regulation 70-25 on the use of volunteers in research.

This material has been reviewed by Letterman Army Institute of Research and there is no objection to its presentation and/or publication. The opinions or assertions contained herein are the private views of the author(s) and are not to be construed as official or as reflecting the views of the Department of the Army or the Department of Defense. *(AR 360-5)*

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COMBAT OCULAR PROBLEMS

Supplement
April 1982

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Letterman Army Institute of Research
Presidio of San Francisco, California 94129

FOREWORD

Since the conference on *Combat Ocular Problems* at Letterman Army Institute of Research in October 1980, researchers in the Division of Ocular Hazards have produced data that indicate new major thrusts in the need for increased emphasis on ocular protection and the requirements for treatment of acute laser injury.

I anticipate that this Supplement to the Proceedings of the Conference on *Combat Ocular Problems* will be of considerable value to Department of Defense agencies who are involved in the research aspects of laser safety as well as implications of the magnitude of ocular injuries which military personnel may sustain.

Current research is a continuation of activities initiated in 1980. Guidance and doctrine will be implemented as new biomedical information are obtained.

JOHN D. MARSHALL, JR.
Colonel, Medical Service Corps
Commanding

April 1982

PREFACE

Within the past year, research in laser bioeffects has been conducted for the wavelengths of Army interest at 800 to 900 micrometers and in the infrared region at 1.732 microns.

Additional experiments have been conducted using neurophysiological and psychophysical evaluations of suprathreshold laser exposures. Also, histological biochemical studies of the effects of Q-switched ruby laser have been performed.

The intent of this Supplement is to provide the user, developer, and safety community the most current data on laser bioeffects. These data should offer them sufficient biomedical information to assist in the evaluation of laser effects (by mathematical analysis) as well as provide additional impetus to the current military program in ocular protection.

EDWIN S. BEATRICE, *M.D.*
Colonel, Medical Corps
Chief, Division of Ocular Hazards

April 1982

ACKNOWLEDGMENTS

Many hours of revision, reorganization and final editing have resulted in the following pages. It is by no means sufficient to applaud the personnel who labored to produce the final product.

To SFC Vernon Farr, whose persistence and friendly persuasion of the authors to make changes and give up final figures for the text, and to SP4 Ardella Edwards, whose long hours on the Composer as well as final proofing of the text assisted so greatly, go individual "Thank you" messages.

And again to a gratious, helpful, supportive technical editor, Lottie Applewhite, who is always of such assistance in making suggestions, organizing and proofing each section, goes a special hurrah.

COMBAT OCULAR PROBLEMS
Supplement April 1982

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Forepages

This group of papers was presented at the 1982 Army Science Conference, June 15-18, West Point, New York. Camera-ready copies were submitted for publication in the Proceedings of that Conference. The group of papers has been collated as a Letterman Army Institute of Research (LAIR) publication for the convenience of our specific user audience.

Stuck, Lund, Beatrice

OCULAR EFFECTS OF RELATIVELY "EYE SAFE" LASERS

Bruce E. Stuck, MS, David J. Lund, BS
and Edwin S. Beatrice, MD, COL MC

Ocular safety is particularly important for laser training devices where multiple devices are utilized simultaneously in two-way exercises to simulate actual engagement scenarios. Visible and near infrared laser systems pose a hazard to the eye at ranges that are tactically significant since the collected radiation is transmitted by the outer ocular medium and focused to a small spot on the sensory retina. For laser wavelengths greater than 2 μm, the cornea absorbs strongly and the ocular response at near threshold doses is confined to the cornea. In the spectral region from 1 to 2 μm, the outer ocular media (cornea, aqueous, lens, vitreous) undergo the transition from highly transparent to essentially opaque. Consequently, the dose required to produce a response and the location of the response within the outer ocular media are dependent on the wavelength. The absorption of the incident radiation throughout a larger volume of tissue results in a higher threshold dose and therefore a reduced ocular hazard.

In previous work in this laboratory, ocular dose-response relationships were experimentally determined for the following wavelengths: 1.33 μm (neodymium), 1.54 μm (erbium), and 2.06 μm (holmium) lasers. In this work, the ocular effects of erbium laser radiation at 1.732 μm were determined for single long pulse exposures (pulse duration of 225 μs). Corneal damage thresholds were determined as a function of corneal irradiance diameters ranging from 500 μm to 1000 μm. The responses determined by biomicroscopy were observed in rhesus monkey eyes. These bioeffects data suggest that a wavelength dependence of the permissible exposure limits be considered for this spectral region. The implications of this research suggest alternatives for laser/laser wavelength selection in the development of "eye safe" lasers for use in Army systems.

* * * * *

Biography of First Author

Present Assignment: Research Physicist, Division of Ocular Hazards, Letterman Army Institute of Research, Presidio of San Francisco, CA.

Past Experience: Research Physicist, Joint AMC-AMRDC Laser Safety Team, Frankford Arsenal, Philadelphia, PA. 1970 - 1974.

Degrees Held: Bachelor of Arts, Catawba College, Salisbury, NC, 1967; Master of Science, Virginia Polytechnic Institute and State University, Blacksburg, VA, 1972.

Stuck, Lund, Beatrice

OCULAR EFFECTS OF RELATIVELY "EYE SAFE" LASERS

Laser devices are an important part of current and future Army systems. Laser rangefinders, designators, communicators, and training devices are currently deployed or are in some stage of development. Most current laser rangefinders and designators, which enhance the effectiveness of the modern Army weapon systems, operate in the visible and near infrared region of the electromagnetic spectrum. The eye is particularly vulnerable in this wavelength region. The collimated laser radiation collected by the eye is transmitted by the ocular media with little attenuation and focused to a small spot on the sensory retina. The retinal irradiance is several orders of magnitude greater than that incident on the cornea; therefore, the total intraocular energy required to produce a retinal lesion is small. Lasers with output characteristic similar to those being fielded are capable of producing serious retinal injury at ranges that are tactically significant (1). The use of binoculars or magnifying optics increases the range at which these injuries can occur. Such devices cannot be used in training exercises without appropriate control restrictions or the use of protective devices. In some cases, training with the actual system in a realistic scenario is inhibited by these restrictions and troop proficiency may never be attained.

Ocular safety is particularly important for personnel using laser training devices where low power laser transmitters and sensitive receivers are used to evaluate the effectiveness of troops and tactics in two-way field exercises which simulate actual engagement scenarios. The MILES (Multiple Integrated Laser Engagement Simulator) program has resulted in the fabrication of laser transmitters configured to simulate several weapon systems. The gallium arsenide laser diodes used in these devices emit near 900 nm. The concern for eye safety when using this system stimulated careful bioeffects research (2) and a continual evaluation of maximum permissible exposures (MPE) given in AR 40-46 and TB MED 279 (3,4). If the emission from a laser system does not exceed the MPE as defined by TB MED 279 (3), then that system is a Class I system and can be referred to as "eye safe." To simulate weapon systems which are effective at longer ranges, lasers which emit more energy per pulse are required to offset losses due to atmospheric absorption and beam divergence. "Eye safe" lasers are desirable for these applications and for rangefinders and designators which can be used without restriction in training exercises. Laser systems operating beyond 1.4 μm have commonly been called "eye safe" and indeed, relative to lasers operating in the visible or near-infrared,

the MPE for direct interbeam viewing is 2000 to 100,000 greater. However, only limited experimental biological effects data exist for wavelengths in this region of the spectrum.

In the spectral region from 1 to 3 μm, the outer ocular structures (cornea, aqueous, lens, vitreous) undergo the transition from highly transparent to essentially opaque. The absorption coefficient varies over 3 orders of magnitude (5). At 10.6 μm, where approximately 90% of the incident energy is absorbed in the first 70 μm of tissue, the corneal response at near threshold doses is confined to the corneal epithelium. Recovery from the insult occurs within 24 to 48 hours as observed by slit lamp microscopy (6,7). As the absorption decreases (in the 1-3 μm region), the incident energy is absorbed and is dissipated over a larger volume of tissue. The absorption of the incident radiation throughout a larger volume of tissue results in a higher threshold dose and therefore a reduced ocular hazard unless deeper structures such as the corneal endothelium or the crystalline lens are more sensitive to the radiation insult. Consequently, the wavelength dependence of the dose-response relationships can be compared to the wavelength dependence of the absorption of the ocular media.

The ocular effects of infrared lasers for specific exposure conditions have been described (2, 6-14). In this paper, experimental ocular dose-response data obtained at 1.732 μm are presented and compared to bioeffects data obtained at other wavelengths in this spectral region.

METHODS

An erbium laser operating at 1.732 μm was fabricated in our laboratory and operated in the long pulse mode. The 1/4 by 3 inch erbium rod (obtained from Sanders Associates, Inc, Nashua, NH) was inserted into an eliptical cavity and pumped by a linear flash lamp (EG&G Inc, FX-42C3, Salem, MA). Energy input to the lamp was approximately 425 Joules. The maximum energy in a single pulse at 1.732 μm was 200 mJ. The emission duration was 225 μs full width half maximum (FWHM) and reached complete extinction at 380 μs. The measured beam divergence was 3.0 milliradian. A schematic of the laser exposure system appears in Figure 1. Because of the limited total output energy, a lens was used to focus the laser energy at the corneal plane. The small amount of energy (100 μJ) transmitted through the highly reflective mirror at the rear of the cavity was proportional to the energy measured at the cornea. Before exposure of the rhesus monkey's eyes, the ratio of the energy at the corneal plane to that at the reference detector was determined. Energy measurements were made with pyroelectric energy monitors (Laser Precision Corporation, Model RkP 335, Utica, NY). These detectors were calibrated with a disc calorimeter (Scientech Model 30-2002, Boulder, CO). Calibrated neutral density filters were placed in the beam to

vary the energy per exposure. The point of intersection of the split beams from a helium neon laser was used to locate the corneal exposure plane and to facilitate selection of the corneal exposure site.

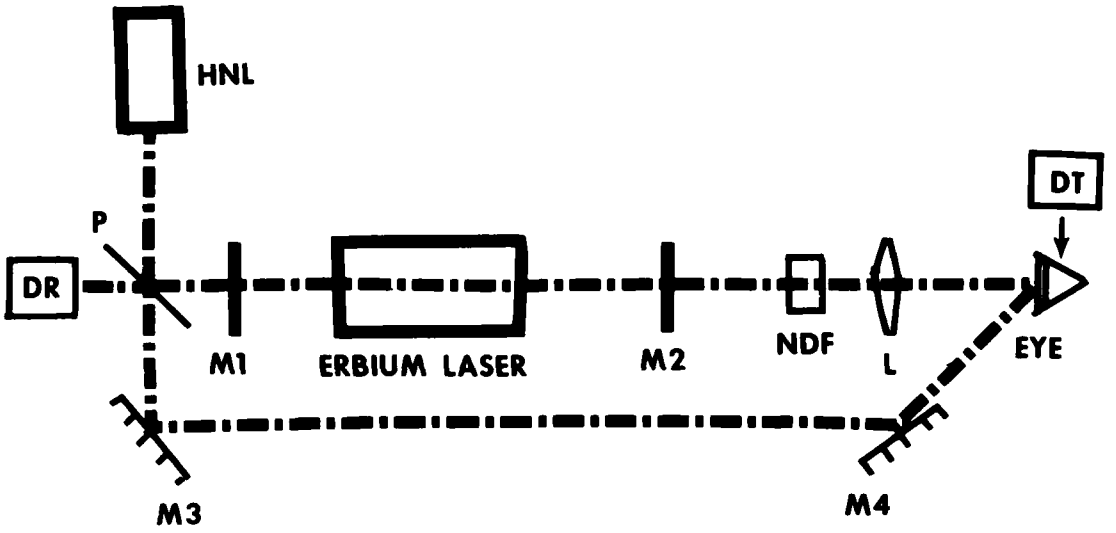

Figure 1. Erbium laser exposure system with emission wavelength of 1.732 μm. DR – Reference detector, DT – Target detector, HNL – Helium neon alignment laser, NDF – Neutral density filter holder, M1 to M4 – mirrors, P – pellicle, L – lens.

Four lenses were used to obtain a range of corneal irradiance diameters. The corneal exposure plane was located in the experimentally determined focal plane a distance of f_p from the lens. The intensity profile of the beam and the effective beam diameter at the corneal plane were measured by two techniques. 1) By systematically reducing the energy per pulse and irradiating developed photographic paper, the relative intensity distribution was displayed. 2) Apertures with progressively decreasing diameters were placed at the exposure plane, and the total energy through each aperture was measured. The intensity profile at the focal plane was "approximately" gaussian and the reported beam diameters ($d_{1/e}$) are the diameters at the 1/e intensity points. The radiant exposure is the peak radiant exposure obtained by dividing the total incident energy by the area defined by the beam diameter ($d_{1/e}$).

Rhesus monkeys (Macaca mulatta) were tranquilized with ketamine intramuscularly and anesthetized with pentobarbital sodium intravenously. The ocular pupils were dilated with one drop each of 2% cyclopentolate hydrochloride and 10% phenylephrine hydrochloride to facilitate biomicroscopic evaluation. The outer ocular structures (cornea, aqueous, lens, and vitreous) were carefully evaluated before

and after exposure by use of the slit lamp biomicroscope. Body temperature during anesthesia was maintained with a thermal blanket. The eyelid was held open with a pediatric eye speculum and the cornea was gently irrigated with physiological saline to prevent drying. Six to nine exposures were placed in each cornea in an array of independent sites (Table 1). The dose was incrementally varied over a preselected range.

Table 1
Corneal ED_{50}s for single 225 μs exposures at 1.732 μm

f_p cm	$d_{1/e}$ μm	ED_{50} (95% CI) J/cm^2	SLOPE	DOSE RANGE TESTED J/cm^2	No. ANIMALS/ EYES/ EXPOSURES
17.8	515	29 (27-31)	1.30	1.0-80	3/6/54
24.1	740	26 (23-29)	1.34	0.5-45	4/7/49
30.5	920	22 (20-23)	1.28	0.3-30	4/8/48
40.6	1200	>16*		14-16	1/2/8

* No ED_{50} was determined for this condition because of the limited energy per pulse available from the laser.

The corneas were evaluated immediately and at 1 hour after the exposure. The response criterion was the appearance of a lesion at the exposure site as observed with the slit lamp biomicroscope. Other evaluations were made at 24 hr, 48 hr, 1 week and up to 6 months after exposure. The crystalline lens was also carefully evaluated. The effective dose for a 0.5 probability of producing an observable response (ED_{50}), the 95% confidence intervals about the ED_{50}, and the slopes of the regression lines through the experimental data (slope = $ED_{84}/ED_{50} = ED_{50}/ED_{16}$) were determined by probit techniques (15).

RESULTS

The ED_{50}s for the production of a corneal lesion at 1.732 μm observed with the slit lamp biomicroscope and the exposure conditions are presented in Table 1. The ocular response for these exposure conditions was confined to the cornea. Corneal lesions generally involved the entire corneal thickness (Figure 2). Lesions near the ED_{50} were smaller and less dense than those produced at 1.5 to 2.0

times the ED_{50}. No lesion was observed at 24 or 48 hours that was not observed at one hour. No lenticular effects were observed at one hour or in the four animals that were evaluated up to 6 months after exposure. Some corneal lesions observed at one hour were not observed at 48 hours. Over the limited range of exposure conditions, the ED_{50} exhibits a dependence on the irradiance diameter (Figure 3). The radiant exposure required to produce a corneal lesion decreases as the irradiance diameter increases.

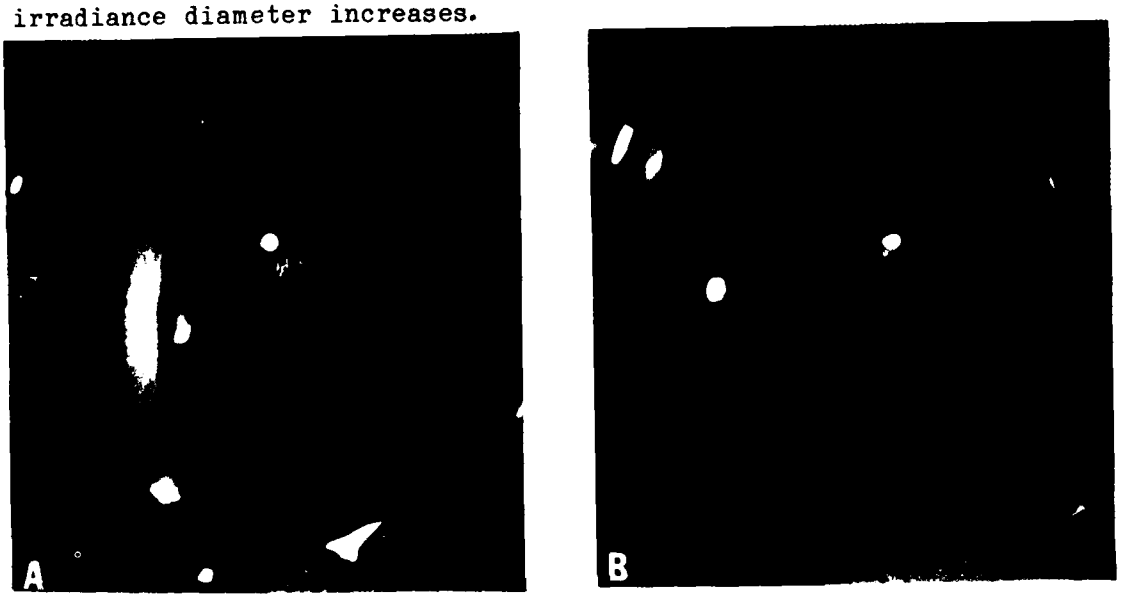

Figure 2. A. Slit lamp photograph of a corneal lesion one hour after exposure produced by an erbium laser operating at 1.732 μm (Corneal radiant exposure = 56 J/cm^2, Exposure duration = 225 μsec (FWHM), Incident beam diameter at the 1/e intensity points = 515 μm).
B. Slit lamp photograph of the same lesion shown in A illuminated with a narrow slit of light showing that the lesion extends through the entire thickness of the cornea.

DISCUSSION

The corneal response resulting from exposure to infrared laser radiation is considered to be the result of a temperature elevation of the tissue. Sufficient energy is absorbed in a finite volume resulting in a localized temperature rise that produces a coagulation or opacification of the medium. Predictive thermal model calculations based on a localized elevation of temperature to a "threshold peak temperature" have been used to estimate the threshold dose required to produce a corneal lesion (16). These thermal model results are considered to be in good agreement with most experimental data published in this wavelength region. Experimental data of this and other experiments are given in Table 2. The ED_{50}s for corneal injury at 1.732 μm are lower than the ED_{50} obtained at the 1.33 μm and higher than those obtained for erbium laser radiation at 1.54 μm. This trend was anticipated based on the relative absorption of cornea at these three wavelengths. Corneal effects at 1.732 μm were similar to those

produced at 1.33 μm and 1.54 μm in that the observed response extended throughout the full corneal thickness.

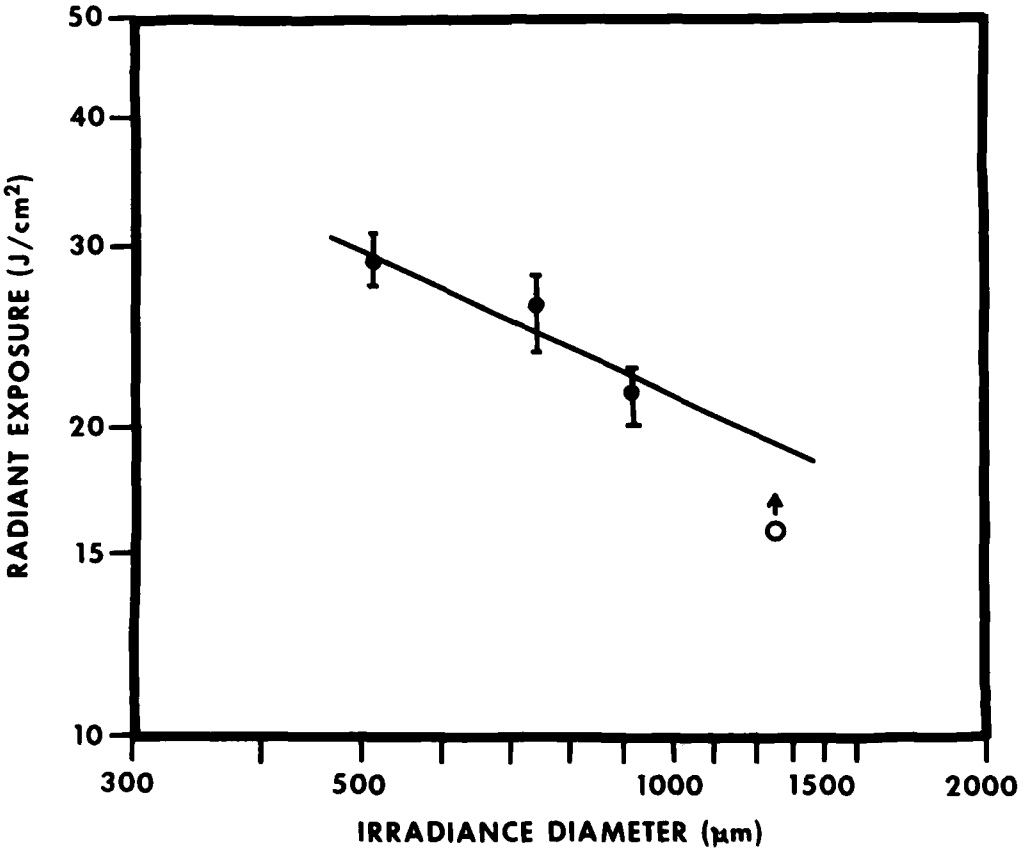

Figure 3. The ED_{50} and 95% confidence interval about the ED_{50} for the production of a corneal lesion as a function of the irradiance diameter of the incident beam ($d_{1/e}$). No corneal effect was observed for exposures made with the 1200 μm irradiance diameter (open circle with arrow).

The ED_{50}s given in Table 2 are plotted in Figure 4 as a function of wavelength to exhibit the wavelength dependence of the damage threshold. Inherent to the wavelength dependence of the ED_{50} is the wavelength dependence of the ocular media absorption. The solid curve on Figure 4 is the depth at which 95% of the incident energy has been absorbed. The absorption coefficients of physiological saline which approximate that of the cornea and outer ocular media were used to calculate the 95% absorption depth. Let x_1 be the depth at which 95% of the incident radiation is absorbed. From Lamberts Law, $I/I_o = e^{-ax_1}$ where I_o is the incident intensity, I is the intensity transmitted through a thickness x_1 of medium with an absoption coefficient of a. By letting $I/I_o = .05$ (i.e. 95% of incident energy absorbed), the depth or thickness x_1 can be calculated for a given absorption

TABLE 2. CORNEAL DAMAGE THRESHOLDS FOR INFRARED LASER RADIATION

WAVELENGTH	EXPOSURE DURATION	IRRADIANCE DIAMETER	CORNEAL ABSORPTION COEFFICIENT[a]	ED_{50}	REFERENCE
μm	s	mm	cm^{-1}	J/cm^2	
1.318-1.338[b]	.25 ms	.40	2.28	45	9
1.41	25 ns	1.1	15.9	2.1-4.2	12
1.54	40 ns	1-2	9.03	4.7	13
1.54	50 ns	1	9.03	21.0	8
1.54	.93 ms	1	9.03	9.6	10
1.54	1.0 ms	1-2	9.03	7.2	13
1.732	.225 ms	.515	5.88	29.0	(c)
1.732	.225 ms	.740	5.88	26.0	(c)
1.732	.225 ms	.920	5.88	22.0	(c)
1.732	.225 ms	1.20	5.88	>16.0	(c)
2.06	42 ns	.32	28.2	5.2	10
2.06	50 ns	.351	28.2	3.25	15
2.06	.10 ms	1.8	28.2	2.9	10
2.6-2.9[d]	45 ns	0.82	>5000	.156	14
2.9	100 ns	10.0	12900	.006-.010[e]	11
3.6-3.9[f]	100 ns	.96	112-180	.377	14
10.6	1.4 ns	10.0	817	.013-.015[e]	11
10.6	100 ns	2.1	817	0.35	2

(a) Corneal absorption coefficients from Reference 5 for wavelengths less than 2.1 um. For wavelengths greater than 2.06 um the absorption coefficient of water which approximates that of the cornea is tabulated.

(b) Multiline neodymium laser with 40% of the energy at 1.318 um and 60% at 1.338 um.

(c) This report.

(d) Multiline hydrogen fluoride laser.

(e) No ED_{50}s was determined. The dose listed is the approximate threshold dose that an immediate response was observed.

(f) Multiline deuterium fluoride laser.

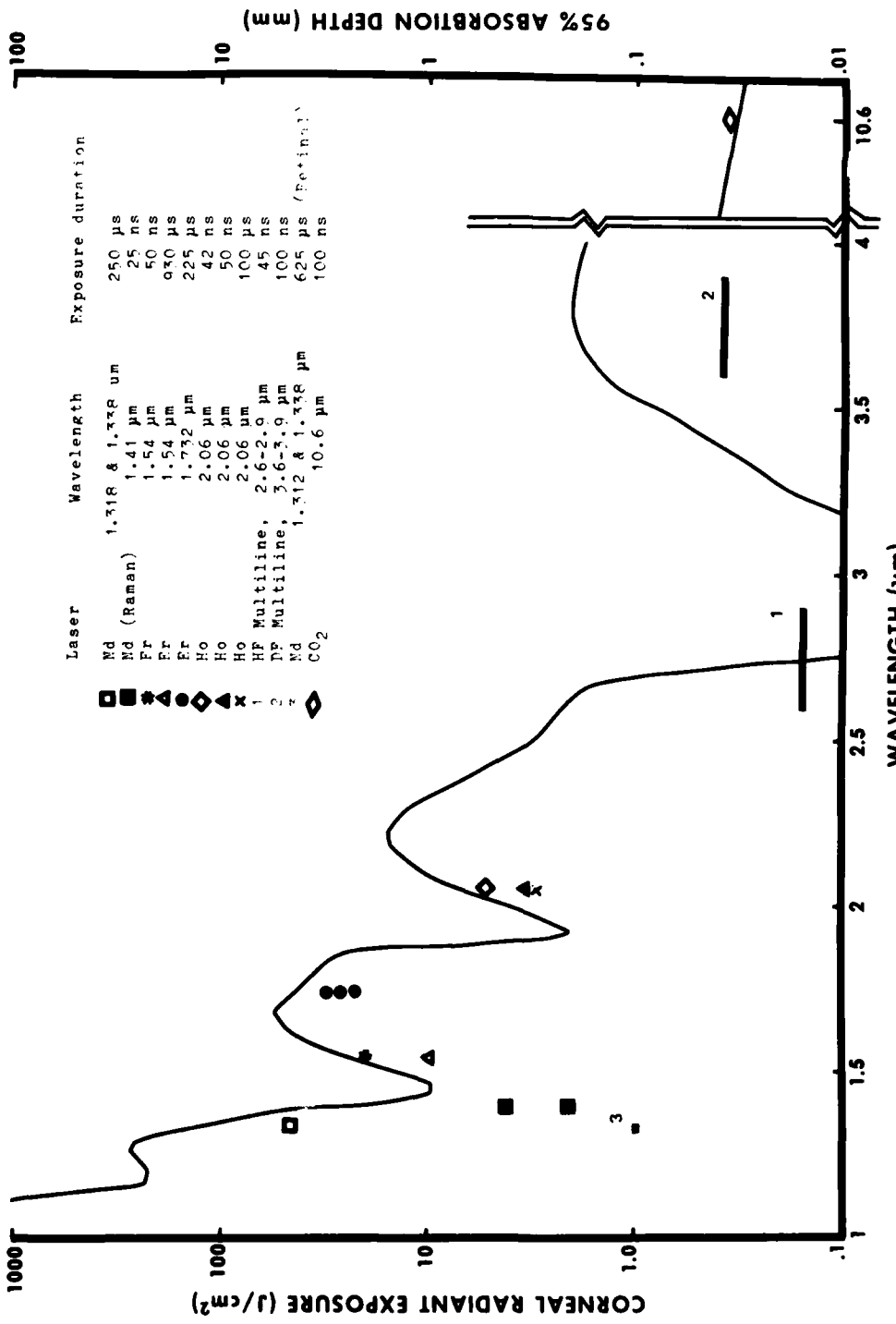

Figure 4. The ED_{95} for the production of a corneal lesion for exposure conditions given in Table 2 as a function of wavelength. The solid curve is the depth (right hand axis) at which 95% of the incident energy is absorbed in physiological saline (the absorption properties of physiological saline approximate those of the outer ocular media). The data point at 3 is the ED_{50} for the production of a retinal lesion obtained at the 1.33 μm.

coefficient a. The volume in which the radiation is absorbed is equal to Ax_1 where A is the cross sectional area of the incident beam. If Q is the incident energy, then the absorbed energy/ unit volume is Q/Ax_1. Assuming the absorbed energy per unit volume required to produce corneal damage is independent of wavelength, therefore $Q/Ax_1 = k$ at the threshold dose where k is a constant. Consequently, the radiant exposure Q/A is directly proportional to the absorption depth or $Q/A = kx_1$. There is a direct correlation between the dose at threshold and the penetration depth (Figure 4).

Even though the exposure conditions (exposure duration, beam diameter), calibration, and observation criteria of different investigators were not identical for the experimental data subjected to this analysis, the wavelength dependence of the corneal ED_{50}s is approximated by the shape of the absorption depth curve. Given identical experimental conditions across investigations and adjustment of absoption depth curve, a better fit to the experimental data may result. Doses required to produce an observable corneal response in the wavelength region between 1 and 2 μm were higher than those required at 2.8, 3.8, and 10.6 μm where absorption takes place within a much smaller volume. The corneal response of a near threshold exposure at the shorter infrared wavelengths involved the corneal stroma and did not exhibit the rapid repair as reported for the longer wavelengths where the threshold response only involved the corneal epithelium. The solid curve in Figure 4 supports that observation. Near threshold lesions at the shorter infrared wavelengths can be considered more severe since a long lasting stromal scar results.

For the exposure conditions evaluated to date at 1.732 μm, no retinal or lenticular effect has been observed; however, further evaluation for a collimated beam continues. The ED_{50} for an ophthalmoscopically visible retinal lesion was establish for the 1.3 μm neodymium laser (2). The beam divergence was 2.3 mr, pulse duration was 650 μs and the corneal beam diameter was 5.5 mm. The total intraocular energy was 356 mJ resulting in a corneal radiant exposure of 1.5 J/cm^2 a the ED_{50}. If this energy were averaged over a 7 mm pupil, the corneal radiant exposure required to produce retinal injury at 1.3 μm is 0.93 J/cm^2. This value is also plotted in Figure 4. At 1.3 μm, the corneal radiant exposure required to produce a retinal effect is much lower (Table 2) than that required to produce a corneal effect; nonetheless, the corneal radiant exposure required to produce a retinal response is 3 orders of magnitude greater than that required at 1.064 μm (2) and the MPEs for both lasers are identical (4).

The dependence of the corneal damage threshold on the irradiance diameter of the incident beam has not been described in previous investigations at any wavelength. Common to many of the investigations of the corneal effects in the infrared has been the necessity to focus the output energy on the cornea (8,9,10,12,13,15)

because of the limited energy per pulse from typical laboratory laser devices operating in this wavelength region. Consequently corneal damage thresholds were obtained only for small irradiance diameters. For irradiance diameters from 500 to 1000 μm, the radiant exposure required to produce a threshold lesion decreased as the beam diameter increased (Figure 2) for 1.732 μm laser radiation. Accidental exposures to infrared lasers will probably involve exposure of the entire cornea (irradiance diameters greater than 10 mm). Further evaluation of this dependence at other wavelengths in this region is required in order that the potential implication to the establishment of permissible exposure limits can be ascertained.

The MPEs for ocular exposure to wavelengths greater than 1.4 μm currently depend only on the exposure duration. These values have been based primarily on the dose-response relationships reported for carbon dioxide laser radiation (10.6 μm). No wavelength dependence of the MPE has been included in laser safety standards. The only exception is the elevated permissible exposures for the Q-switched erbium laser (1.54 μm) where experimental data (5) existed when these permissible exposure limits were established (3). The MPE for a single exposure less than 100 μs in duration is 10 mJ/cm^2 for laser radiation with wavelengths greater than 1.4 um (1 J/cm^2 for 1.54 μm radiation). The MPE for ocular exposure to laser radiation at 1.732 μm or 2.06 μm is the same as the MPE at 10.6 μm, even though the ED_{50}s differ by a factor of 10 to 100. Although additional experimental dose-response data are needed in the 1 to 3 μm region for longer exposure durations, larger corneal irradiance diameters, and repetitive pulse conditions, a generalized wavelength correction to the MPE in the infrared spectral region is indicated by these experimental data. When compared to the MPEs for visible and near infrared radiation (the MPE ranges from 0.5 to 5 $\mu J/cm^2$ for exposure durations less than 100 μs), lasers operating beyond 1.4 μm are relatively "eye safe." Lasers operating in the IR-B region which emit 100 mJ per pulse could be used without stringent range control restrictions or protective devices. With current permissible exposure limits, a 1.54 μm laser would be desirable since the MPE is 100 times that for other systems such as holmium (2.06 μm) or erbium (1.732 μm).

CONCLUSIONS

Ocular dose-response data obtained at 1.732 μm for exposure conditions examined thus far coupled with the other experimental data obtained in the wavelength region from 1.3 to 3.0 μm support consideration of including a wavelenght dependence in the maximum permissible exposure. This wavelength dependence should be based on the relative absorption properties of the ocular media. Lasers which operate in this wavelength region offer a distinct advantage to the system developer from an "eye safety" standpoint.

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Lund, Beatrice, Schuschereba

BIOEFFECTS CONCERNING THE SAFE USE OF GaAs LASER TRAINING DEVICES

David J. Lund, BS, Edwin S. Beatrice, MD, COL MC
and Steven T. Schuschereba, MA

Guidance for the safe use of lasers is provided by AR 40-46 and TBMED 279 in terms of the maximum permissible exposure (MPE). Historically, the MPE has been derived from acute bioeffects data, most commonly the ED_{50}s for the production of an ophthalmoscopically visible retinal alteration when the laser beam is collimated to irradiate a minimum retinal area. The ED_{50} is defined as that dose which has a 50% probability of producing an alteration. TBMED 279 therefore reflects the accuracy and completeness of the bioeffects data base. Until recently, no laser bioeffects data existed in the spectral region between 694.3 nm (ruby laser) and 1060 nm (neodymium laser). Lasers which operate in this spectral region include the gallium arsenide (GaAs) semiconductor lasers widely employed in training devices, the use of which necessitates a high probability of direct exposure of personnel. The MPE in this spectral region is obtained by interpolation between the MPEs for 694.3 nm and 1060 nm. Recent measurements of the ED_{50} for retinal alteration at 850 nm (erbium laser) indicate that the interpolation is not accurate, but provides a MPE which is higher than the bioeffects data would indicate. This result has led to a LAIR effort to determine accurately the wavelength dependence of the ED_{50} within the wavelength range from 850 to 900 nm using a pulsed dye laser as the irradiance source. The data of this experiment show that the ED_{50} is as much as an order of magnitude lower than the projection based on the interpolation from 694.3 nm to 1060 nm. However, the provisions of TBMED 279 do provide a safety margin of 10 in this spectral region.

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Biography of First Author

Present Assignment:	Research Physicist, Letterman Army Institute of Research
Past Experience:	Research Physicist, Joint AMC-AMRDC Laser Safety Team and Laser Countermeasures Division, Frankford Arsenal, Philadelphia, PA, 1961-1975.
Degrees Held:	Bachelor of Science, Western Illinois University, Macomb, Illinois. 1961. Graduate studies in physics, Temple University, Philadelphia, PA, 1964-1972

Lund, Beatrice, Schuschereba

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Modern weaponry poses a vexing problem to the military training community: how can realistic battlefield games be conducted which mimic the undeniable ability of weaponry to produce lethal action at a distance? A child's pointed "BANG, you're dead!" suffers in that it has an unverifiable effect, is limited in range, and exposes the originator to immediate retaliation by the target. It does, however, contain the essential ingredients of a weapons simulation system; the ability to engage a target and transmit a message which can be interpreted by the target in terms of its subsequent ability to function. A key aspect of the message is that it implies a fatal hit but does not possess that element of real lethality.

Systems have been devised which simulate the firing of live munitions for training purposes. The MILES system, developed by PM TRADE, is an example. A gallium arsenide (GaAs) laser transmitter is mounted on, and boresighted with, each weapon, and all potential targets are equipped with detectors sensitive to GaAs laser radiation. When a weapon is triggered, no projectile is fired; rather a signal is transmitted from the laser and directed at the intended target. The success of the round is scored upon receipt of the signal at the target. The system is effective; however it does present a new problem. The MILES system transmits laser beams at personnel; the probability of their eyes being exposed is high. It is essential that the consequences of such ocular exposure be understood to insure that the signal used for simulation does not carry potential harm.

Within the Army, safety restrictions on the use of all lasers are governed by the provisions of laser safety standards, AR 40-46 and TBMED 279 (1,2). In a quest for effectiveness, the designers of MILES have pushed the emitted power to the maximum permissible exposure (MPE) allowed by the standards. An understanding of the potential hazard of the MILES system can be gained by testing the accuracy of the provisions of the safety standards.

TBMED 279 dictates the MPE for laser viewing as a function of several laser parameters including wavelength, exposure duration, effective irradiance diameter and repetitive pulse factors. The relative values of the MPE were derived from bioeffects data which related the potential for tissue alteration to the operational characteristics of the laser. The eye is the most vulnerable part of the body to visible and near infrared laser radiation. This is true

because within the eye a light absorbing layer of tissue (retina) lies at the focus of the eye lens system. Just as the rays of the sun can be concentrated by a lens to burn wood, so is a laser beam concentrated onto the retina where the concentrated energy can induce thermal, mechanical, and chemical processes which alter the retinal tissue.

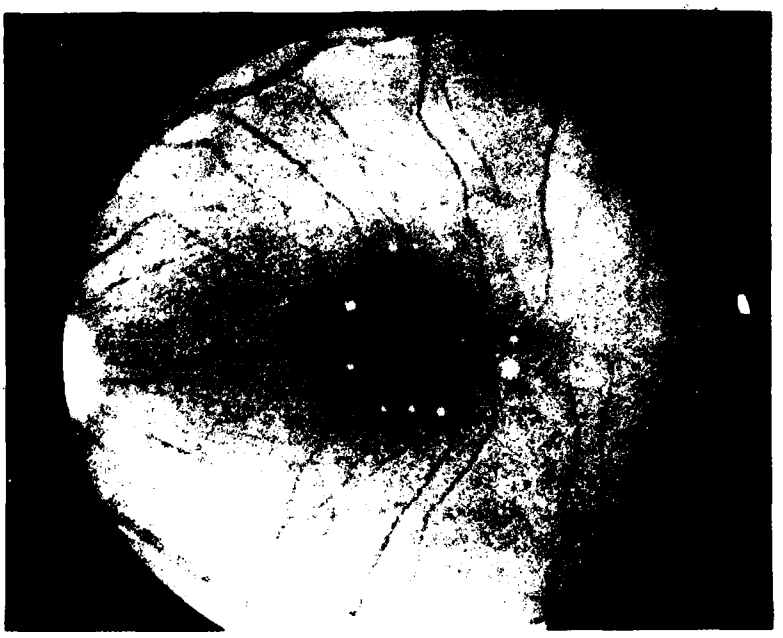

Figure 1. Rhesus monkey retina with laser-induced damage.

The MPE has, for the most part, been based upon the ED_{50} for visible retinal alteration in rhesus monkeys under a given set of exposure conditions. What does that mean? Figure 1 is a photograph of the retina of a live rhesus monkey. The retina is the thin layer of tissue at the back of the eye which contains the visual photodetectors. Damage to the retina can diminish vision. The relatively dark circle near the center is the macula, the area of central vision, and in the center of the macula lies the fovea, the area of most acute vision. Around the periphery of the macula in this photograph are a series of small white spots. These are alterations to the tissue caused by laser exposure. The criterion for laser induced damage, in studies upon which MPEs are based, is the appearance of such a visible alteration. The retina is not uniform in appearance but exhibits large and small scale variations in pigmentation. Because of this variation, the proportion of an incident laser beam absorbed by the tissue will not be the same for all exposure sites. If a series of retinal sites are subjected to laser exposure all at the same incident energy, not all will exhibit the same effect. Some sites will show visible alteration; some will not. For a given incident energy level, there will be a probability

of alteration, computed by dividing the number of exposures producing alteration by the total number of exposures. When such an experiment is performed for a number of exposure levels, a curve is derived which relates the probability of alteration to the exposure level. The probability is low for low level exposures and high for higher level exposures. The data relating the probability of damage to the exposure energy can be processed by the statistical technique of probit analysis (3) to determine the incident energy having a fifty percent probability of producing alteration. This incident energy, or dose, is called the ED_{50}. The ED_{50} is not a damage threshold but rather a statistical point which has greater confidence than any other point on the dose-response curve. The MPE is the maximum permissible exposure for safe viewing of laser radiation. No alteration should ever occur upon exposure at the MPE, which therefore must be lower than the ED_{50}. Based on a number of considerations, the MPE has been chosen to be a factor of 10 to 100 below the ED_{50} upon which it is based. The variation of safety factor results from simplification of the dependence of MPE upon exposure parameters.

The most recent version of TBMED 279 was issued in 1975. At that time essentially no laser bioeffects data existed for the spectral range between 694.3 nm (ruby laser) and 1060 nm (neodymium laser). Faced with the absence of data, the writers of the standards chose to compute the MPE in this range by interpolation between the MPEs at 694.3 nm and 1060 nm. The interpolation was not arbitrary but was based on the transmission and absorption properties of ocular tissue. Figure 2 shows the relationship between the MPE and wavelength for the visible and near infrared. The ED_{50}s are also shown for some specific laser wavelengths, valid for ocular exposure to single short duration pulses. The MPE and ED_{50} are given in terms of total intraocular energy (TIE), that is, the total energy entering the eye. If the MPE is to be an accurate derivation of the ED_{50} for wavelengths between 694.3 nm and 1064 nm, the ED_{50} should closely follow a straight line between these wavelengths. The ED_{50} for 850 nm (erbium laser), recently obtained at LAIR (4), lies significantly below the expected value. Thus we have doubts about assumptions underlying the interpolated MPE in this spectral region. In light of this evidence, it became urgent to determine the wavelength dependence of ED_{50} near the GaAs wavelength of 900 nm.

Advances in dye laser technology have made this study possible. The lasing medium of such a laser is a fluorescent dye carried in a suitable liquid solvent. When optically pumped with intense radiation from a flashlamp or laser, the dye can be caused to emit laser radiation at any wavelength within its fluorescent spectrum. The fluorescent bandwidth of most dyes is nanometers wide, and dyes are available which allow selection of any wavelength from the ultraviolet to greater than 900 nm.

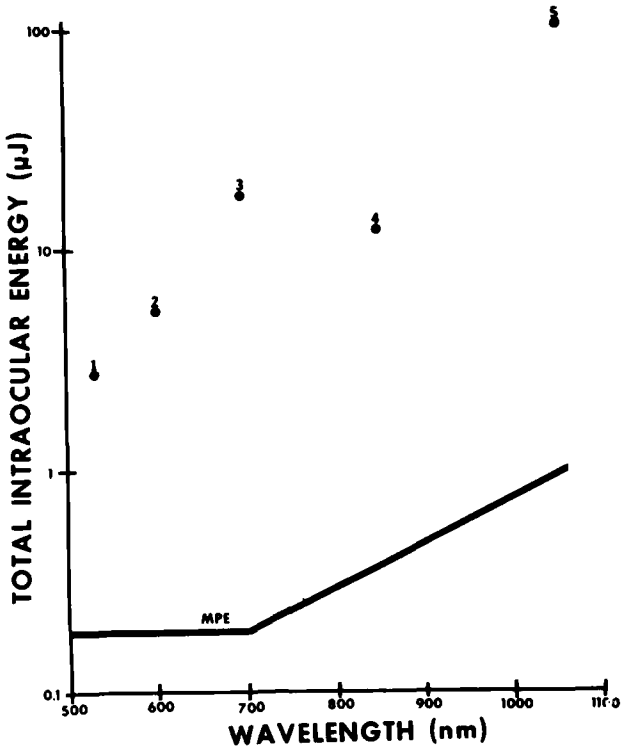

Figure 2. Wavelength dependence of the maximum permissible exposure (MPE) and the ED_{50}s for retinal damage in the rhesus monkey. Given ED_{50}s are for: 1 – 532 nm, 140 ns 4 – 850 nm, 180 ns
2 – 600 nm, 400 ns 5 – 1064 nm, 180 ns
3 – 694.3 nm, 15 ns

The experiments and results of our studies with eyes of rhesus monkeys are reported in this document.

MATERIALS AND METHODS

The source of laser radiation in this study was a Molectron DL-18 dye laser coupled to a Molectron MY33 Nd:YAG laser. The neodymium laser emitted 15 ns duration pulses at a repetition rate of 10 Hz and was provided with internal second and third harmonic generators which could be positioned so that the laser output was any of three wavelengths: 1064 nm (fundamental), 532 nm (second harmonic), or 355 nm (third harmonic). The output energy of the neodymium laser served as an excitation source for the dye laser which consisted of a side-pumped cell through which the dye was circulated, a diffraction grating which served as a wavelength tunable resonator mirror, and an output resonator mirror. The output wavelength of the dye laser was determined by the grating with the restriction that it be tuned to a wavelength within the fluorescent spectrum of the dye. The fluorescent spectrum of the dye in turn was dependent on the specific dye chosen, the dye solvent and concentration, and the excitation wavelength.

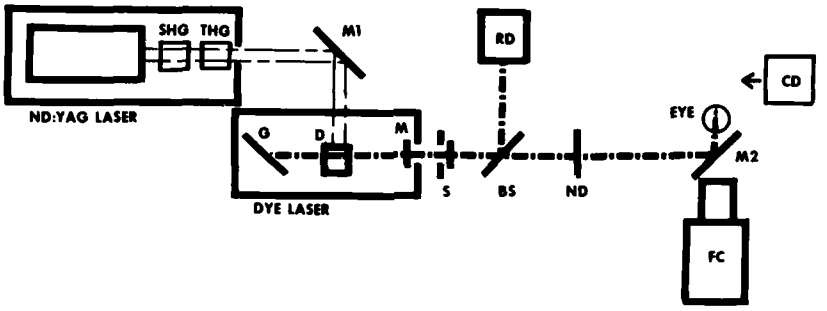

Figure 3. Exposure configuration.
BS – beamsplitter CD – calibrated detector for dosimetry D – laser dye cell
FC – fundus camera G – diffraction grating for dye laser tuning
M – dye laser output mirror M1 – redirecting mirror for Nd:YAG beam
M2 – dichronic mirror ND – neutral density attenuating filter
RD – reference detector S – shutter SHG – second harmonic generator
THG – third harmonic generator

Figure 3 is a schematic of the exposure system. The laser emitted a continuous train of pulses at 10 Hz; a shutter allowed selection of a single pulse for exposure. A beam splitter deflected a constant proportion of the pulse energy into a reference detector while the remainder of the energy passed through attenuators and onto a dichroic mirror. The mirror had high reflectivity at wavelengths longer than 700 nm but was transparent in the visible. A fundus camera, looking through the mirror, permitted observation of the retina to be exposed. The fundus camera, mirror, and laser beam were aligned so that the laser energy reflected by the mirror passed through the center of the ocular pupil and struck the retina at the site corresponding to the crosshairs of the fundus camera viewing optics. Before the rhesus monkey was positioned, a calibrated detector, which directly read the incidence pulse energy, was placed so that it would receive all the energy that would normally enter the eye. The ratio of the energy at this position to the energy at the reference detector was obtained with the attenuator removed. Subsequently, when the eye was exposed, the energy entering the eye for each exposure was determined by multiplying the energy at the reference detector by the ratio previously determined and by the transmission of the attenuating filter chosen to give the desired energy. The laser wavelength, beam divergence, and pulse duration were determined for each wavelength. A Jarrell-Ash 1/2 meter spectrometer was used to measure the wavelength. The wavelength scale of the spectrometer was calibrated against a mercury spectral source; the subsequent laser wavelengths error was less than 0.1 nm. The beam divergence was measured by a linear detector array at the focal plane of a one meter lens.

Rhesus monkeys were used in this study. Each animal was sedated and anesthetized for exposure, its pupils were dilated, and the eye to be exposed was held open by a lid speculum. While the eye was open, the cornea was periodically irrigated with physiological saline solution to maintain clarity. For each test, an animal was positioned and 30 exposures were placed in an array in the extramacular retina. The initial exposures in each sequence were at a dose high enough to produce an immediate visible tissue response. Subsequent exposures were at successfully lower doses so that the range of doses in the array varied by about a factor of ten. The retina was photographed and the exposure sites marked on the photograph for subsequent identification. The exposure sites were examined by ophthalmoscope one hour after exposure and the presence or absence of visible alteration noted for each site. The response at each site was correlated to the dose at that site. For each wavelength, the data obtained by exposure of four to six eyes were statistically evaluated to determine the ED_{50} and associated 95% confidence limits. One animal, exposed to 900 nm radiation, was sacrificed one hour after exposure and the retinas prepared for histological evaluation.

RESULTS

The ED_{50} for single Q-switched exposure was determined for six laser wavelengths obtained from the dye laser. The wavelengths and exposure conditions are listed in Table 1. The solvent for all the laser dyes was DMSO. The laser linewidth at 912 nm was 0.4 nm. For the other wavelengths the laser linewidth was less than 0.03 nm, the resolution limit of the monochrometer used for wavelength measurements. The laser beam was nearly gaussian in profile. The beam divergence was measured at the diameter where the intensity fell to 1/e times the peak value.

The ED_{50} for visible retinal alteration at one hour in rhesus monkey is given in Table 2 for each of these wavelengths. Also given are the 95% confidence limits about the ED_{50} and the slope of the regression line, defined as ED_{84}/ED_{50}. The data are for extramacular exposure.

Table 1
Dye laser parameters

WAVELENGTH (nm)	PULSE DURATION (nsec)	BEAM DIVERGENCE (mrad)	DYE *	CONCENTRATION (molar)	EXCITATION WAVELENGTH (nm)
850.2	11	1.4	DTTC HIDC†	1.5×10^{-3} 1.1×10^{-3}	532
859.6	10	1.6	IR144	6×10^{-4}	532
867.0	7	1.6	IR144	6×10^{-4}	532
880.0	14	1.4	IR125† HIDC†	1.8×10^{-4} 1.8×10^{-3}	532
899.7	6	1.6	IR125	2×10^{-3}	355
912.0	7	1.6	IR140	3×10^{-3}	532

* Dyes listed are products of Exciton Corporation, P.O. Box 3204, Overlook Station, Ohio 45431.
† Two dyes were used in combination; concentration of each is listed in column to right.

Table 2
Retinal ED_{50} for Q-switch dye laser exposure in rhesus monkey

WAVELENGTH (nm)	ED_{50} (μJ)	95% LIMITS (μJ)	SLOPE
850.2	9.1	7.8-10.7	1.71
859.6	6.7	5.6-8.0	1.77
867.0	5.2	4.4-6.0	1.69
880.0	6.3	5.2-7.7	1.80
899.7	4.3	3.4-5.3	2.38
912.0	5.5	4.6-6.7	1.87

Histologic evaluation

Retinal tissue from two eyes exposed to 900 nm radiation was processed and sectioned for light microscopy. Figure 4 is a retinal photograph of one eye taken just prior to sacrifice of the animal. Sites marked 1, 2, 3 and 5 were each exposed to a train of 100 pulses at 10 Hz. The energy per pulse in the train was 17 μJ. Between sites 1 and 2 and between sites 3 and 5 were placed 6 exposures, each consisting of a single pulse having an energy of 17 μJ. Figure 5 shows a section through one of the extramacular sites exposed to 100 pulses. The sensory retina is a complex tissue within which have been defined a number of layers. The retinal pigment epithelium (RPE) is a single layer of cells which contain the pigment melanin. Melanin is the strongest optical absorber in the retina; thus for most laser wavelengths, the RPE is the center of damage. Each photoreceptor of the retina extends through four layers, the outer segment layer (OS), the inner segment layer (IS), the outer nuclear layer (ON), and the outer plexiform layer (OP). The outer segment of the photoreceptor contains the photochemicals which convert the optical signal to a bioelectric signal, the inner segment and nucleus contain the life support system of the cell, and a nerve process extends into the outer plexiform layer where the bioelectrical signal is passed to other nerve cells which convey the information to the brain. Figure 5 shows that, although the damage to the RPE is slight, the photoreceptors at the exposure site have been damaged throughout their length.

The retina contains two types of photoreceptors; the rods which respond to dim light, and the cones which respond to high ambient light and provide color vision. The two types of photoreceptors are not uniformly distributed in the retina: cones are more common in the macula and rods are more common in the extramacular retina. Figure 6 shows a lesion near the edge of the macula where both rods and cones are found. The dose producing this lesion was the same as that for Figure 5. The RPE is more extensively damaged in this lesion, and again the photoreceptors are damaged throughout their length. Figures 7 and 8 are magnified views of the lesions of Figure 5. It can be seen that although the rods are extensively damaged, the cones are relatively unaltered.

DISCUSSION

When the ED_{50} is plotted as a function of wavelength (Figure 9), a minimum is seen near 900 nm. This is difficult to explain on the basis of the known optical qualtities of the rhesus eye. Incident radiation must be absorbed by tissue to produce an alteration. A laser beam entering the eye passes through the cornea, aqueous, lens, and vitreous before reaching the retina. These transparent media absorb a fraction of the incident radiation. Of the radiation reaching the retinal surface, part is reflected and part transmitted through the retina. The remainder of the radiation on the surface is

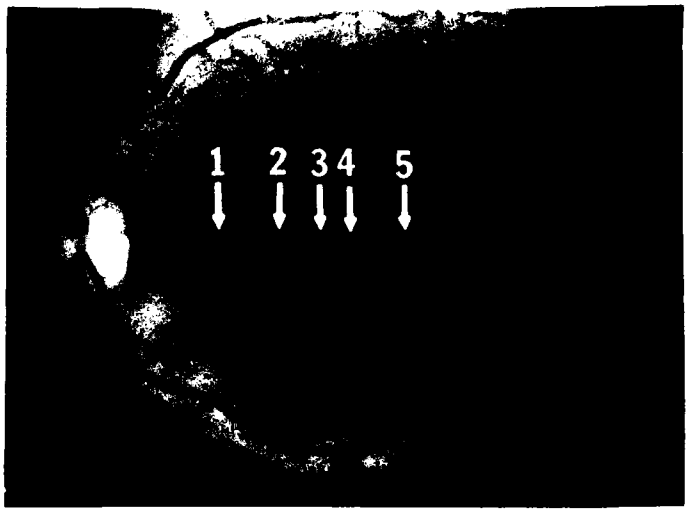

Figure 4. Photograph of rhesus monkey retina. The large white area is the optic disc and the dark area the macula. Tiny white spots (arrows) are focal 900 nm dye laser lesions. There are six barely visible lesions between arrows 1 and 2 and between arrows 3 and 5. Arrow 4 points to one of the lower level lesions.

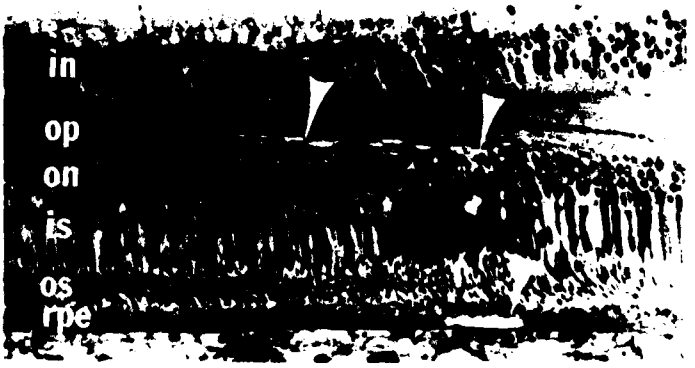

Figure 5. Light micrograph of lesion 2 of figure 1. The section shows the following layers of the retina: inner nuclear layer (IN); outer plexiform layer (OP); outer nuclear layer (ON); photoreceptor inner segments (IS); photoreceptor outer segments (OS); retinal pigment epithelium (RPE). Small arrows indicate vacuolization. Dark stained nuclei are present in the RPE and ON. Some of the outer segments above the lesion are highly swollen. BAR = 100μm.

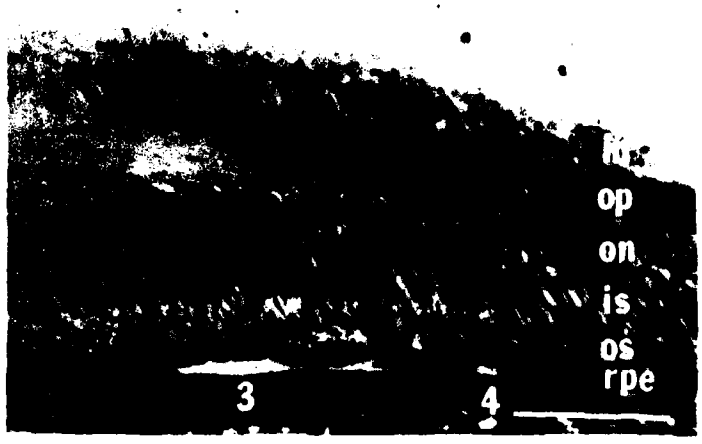

Figure 6. Light micrograph of lesions 3 and 4. Large vacuoles are present in the basal region of the RPE in both lesions and small vacuoles are present in the ON. Dark staining nuclei appear in the RPE and the ON. BAR = 100μm.

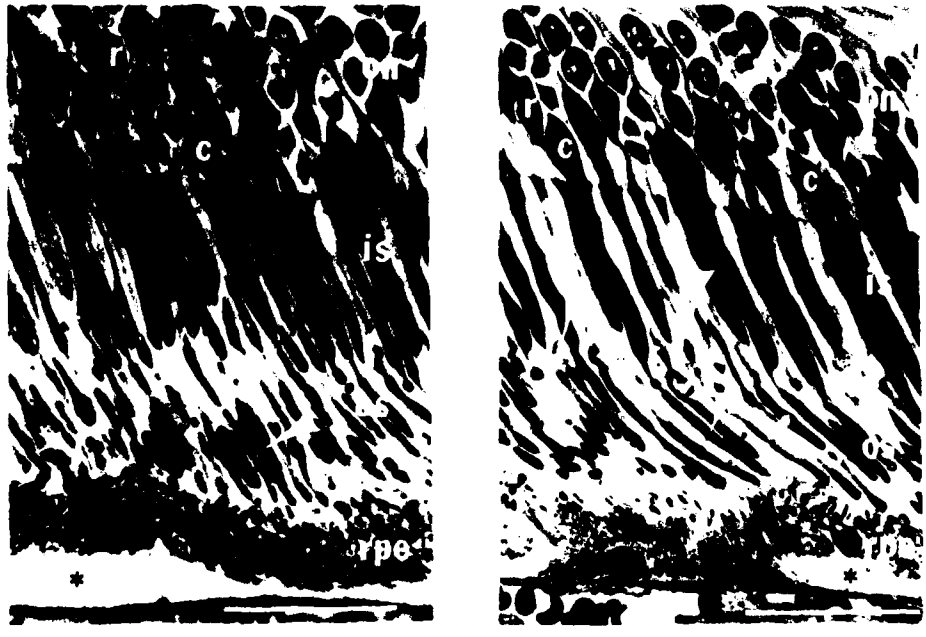

Figure 7 (left lower). Light micrograph of lesion 3. A large vacuole (*) is seen in the basal region of the RPE. Melanin granules are clumped and disarrayed in this layer. Dark staining nuclei are present in the RPE as well as in the ON. Dark stained nuclei belong to rods (r), while cone nuclei (c) stain normally. The slender inner segments of rods are swollen and vacuolated while the larger cone inner segments are normal. BAR = 50μm.

Figure 8 (right lower). Light micrograph of lesion 4. A vacuole (*) is present in the basal region of the RPE. Dark stained nuclei are present in the basal region of the RPE and one dark nucleus is present in the ON. The dark nucleus belongs to a rod (r) that has its outer segments near the lesion in the RPE. Inner segments of rods are highly swollen and vacuolated. The most severe vacuolation occurs at the junction of IS and OS (large arrow) adjacent to the high energy lesion. The cone (c) outer segments show whorl formation about midway in their lengths. BAR = 50μm.

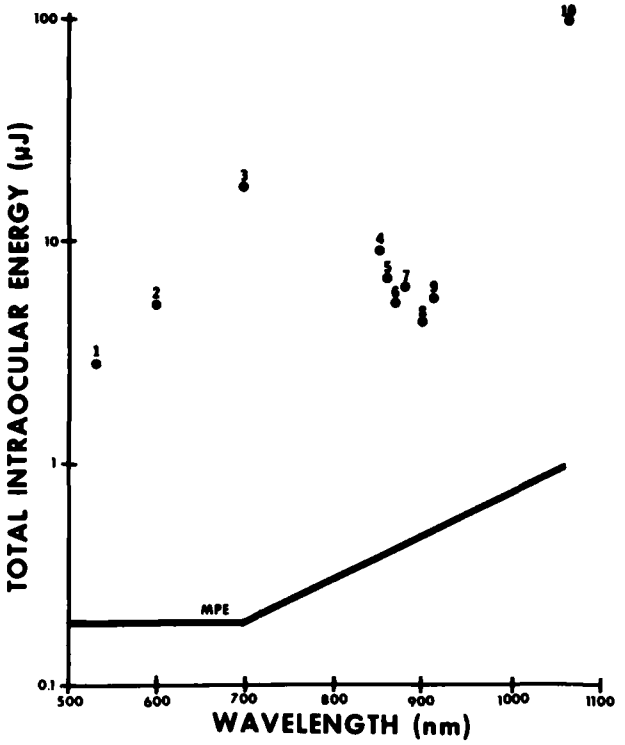

Figure 9. Wavelength dependence of the MPE and the ED_{50} for retinal damage in the rhesus monkey. Given ED_{50}s are for:

1 – 532 nm, 140 ns	6 – 867 nm, 7 ns
2 – 600 nm, 400 ns	7 – 880 nm, 14 ns
3 – 694.3 nm, 15 ns	8 – 899.7 nm, 6 ns
4 – 850.2 nm, 11 ns	9 – 912 nm, 7 ns
5 – 859.6 nm, 10 ns	10 – 1064 nm, 180 ns

absorbed in the retinal tissue. Absorption and reflection by the ocular media are wavelength dependent quantities which have been measured by several investigators (5-7). From these data, the energy absorbed in the retina at each wavelength can be computed as a fraction of the energy incident at the cornea. A transformation of this relationship provides the relative energy at the cornea as a function of wavelength to produce constant absorbed energy in the retina (Figure 10). Comparing this result to the bioeffects data for 850 nm to 900 nm, we are driven to consider two possibilities: either there is a flaw in the ocular absorption measurements; or laser radiation in this spectral range produces retinal alteration with lower absorbed energy. The optical absorption measurements of the rhesus eye were performed in vitro with low level illumination. These measurements would not detect absorption mechanisms which operate only in living tissue nor would they detect power dependent interactions. Photochemical interactions, normally associated with shorter

wavelengths and longer exposure durations, requi e lower energy for initiation than do the thermal interactions normally associated with near infrared retinal alteration. The retina is a beehive of photochemical and biochemical processes which might be altered by specific wavelengths. The histopathology suggests that 900 nm radiation is absorbed by the rod photoreceptors to the extent that the underlying RPE is relatively spared in the rod-rich areas of the retina. The data currently available are not sufficient to explain the reduced ED_{50} near 900 nm.

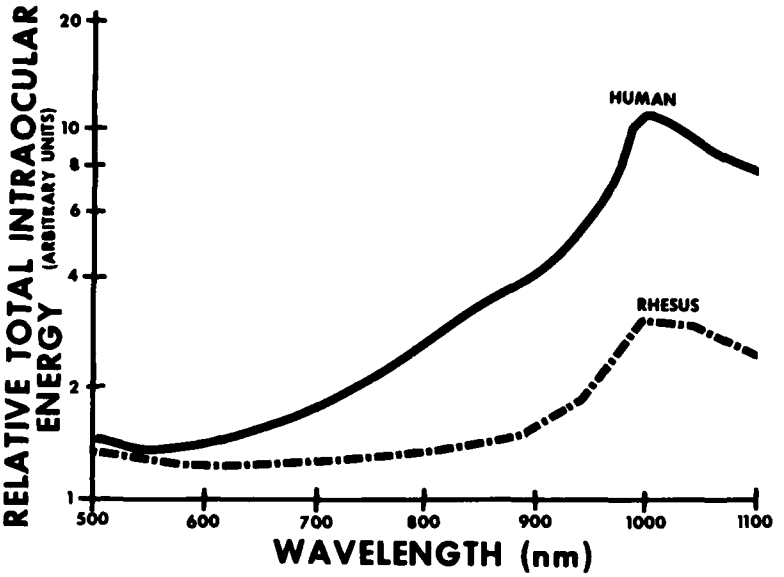

Figure 10. Wavelength dependence of the total intraocular energy required to produce a constant level of absorbed energy in the retina of human and rhesus monkey eyes.

The determination of ED_{50}s is not an exact science; the shape of the ED_{50} versus wavelength relationship from 700 nm to 1060 nm may not be exactly represented by the data of Figure 9. However, the data are internally consistent in that all the data were collected at LAIR by the authors and have a commonality of dosimetry and determination of results. All the ED_{50}s of Figure 9 are for single short pulse exposure to the extramacular retina in rhesus monkey. There is a real decrease in the ED_{50} between 850nm and 900 nm as compared to the ED_{50}s at 700 nm and 1060 nm. The MPE as provided in TBMED 279 is not consistent with these new bioeffects data. When the MPE at 900 nm was compared to the data of this experiment, we found a safety factor of 10. The current MPE should provide adequate protection for exposure to single pulses from the GaAs laser.

CONCLUSIONS

The MPEs for wavelengths affecting GaAs laser training devices were derived based on assumptions concerning interaction of laser radiation with retinal tissue. The data of this study show that those assumptions were not adequate, and that retinal damage occurs at lower doses than expected. The results of this study do not necessarily have an adverse affect on the use of GaAs laser training devices - the safety standards still provide an adequate safety margin. The bioeffects data base in this spectral region is still incomplete; additional data now being collected will serve to define the wavelength dependence of ED_{50} between 700 nm and 1060 nm. It is possible that, when the data base is complete, a recommendation will be made to change the operating wavelength of training devices to a wavelength with increased safety margin. The data currently available do not warrant a recommendation for modification of the safety standards.

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Randolph, Schmeisser, Beatrice

LASER FLASH EFFECTS: A Non-Visual Phenomenon?

David I. Randolph, PhD, Elmar T. Schmeisser, PhD, CPT MS
and Edwin S. Beatrice, MD, COL MC

Steady state visual evoked potentials (VEPs) were recorded from cynomolgus monkeys in response to an oscillating grating. Single 20 nsec Q-switched ruby laser pulses were directed into the fovea of the experimental eye during the pattern stimulation. No immediate effects on the VEP were noted. Strong delayed effects occurred 45 to 120 sec post-exposure. These effects included large phase shifts in the response signal as referenced to the stimulus, magnitude decreases, variance increases and a loss of waveform correlation with pre-exposure baseline signals. These effects persisted for approximately 30 sec before the VEP re-entrained itself and again demonstrated a normal conformation. Several explanations for these findings are proposed. First, vision is minimally affected by flashes at this wavelength. Second, the 20 nsec pulse may be too fast to produce a measurable response. The delayed effects were consistent with the timecourse of the development of retinal edema. The subsequent recovery of the VEP to its normal synchronization implies that the phenomenon may be non-visual.

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Biography of First Author

Present Assignment: Research Psychologist, Division of Ocular Hazards, Letterman Army Institute of Research, Presidio of San Francisco CA 94129

Past Experience: Research Psychologist, Joint AMC-AMRDC Laser Safety Team, Frankford Arsenal, Philadelphia, PA. 1970-1974

Degrees Held: B.A., Northeastern Univ. Boston MA, 1960
M.A., Northeastern Univ. Boston MA, 1962
Ph.D., Univ. of Massachusetts, Amherst MA, 1965

Randolph, Schmeisser, Beatrice

LASER FLASH EFFECTS: A Non-Visual Phenomenon?

Ruby and neodymium laser rangefinders, ground locator-designators and other devices which emit short (20 nsec) high energy flashes of laser radiation are currently being deployed to troop units in the field. Evidence of the effects of short laser pulses delivered in known quantities and spot sizes on the human retina has been limited to the treatment of proliferative diabetic retinopathy or other clinical states which involve abnormal ocular conditions(1). In these cases, the bulk of the laser energy is directed to the peripheral retina and, when necessary, to the capillary-free zone of the macula. No exposures are placed in the central fovea where visual acuity is best. The soldier using binoculars or other optical sighting devices in the combat environment would receive a laser flash directly in the fovea.

Research on flash effects with human subjects has been generally limited to white light, non-laser sources with large retinal spot sizes (2). The immediate effects upon the vision of individuals who receive foveal laser exposures (minimal spot size) is unknown. It is thus important to be able to predict accurately the biological and functional effects of these exposures, delineate the physical and physiological parameters and recommend a course of treatment for those thus exposed. Ultimately, these data should lead to techniques for preventing debilitating laser bioeffects.

Laser energy levels, wavelength, size of the affected area, pulse length, pulse repetition rate and other physical variables have been related to changes in the eye and skin since the mid 1960s (3,4)and have been primarily concerned with both gross and microscopic alterations in the tissue. Based upon these changes, inferences have been made about the functional effects, i.e. a lesion in the retina implied a loss of vision at that site.

In order to quantify the implied loss of vision, nonhuman primates have been trained to respond to acuity criteria for high and low stimulus contrast targets. Robbins et al (5) reported immediate (within 2 min) high target contrast visual acuity decrements in the rhesus monkey following foveal exposure to 100 msec pulses of helium-neon (633 nm), krypton (647 nm) and argon (514 and 488 nm) laser lines. The spot sizes varied between 150 and 300 μ. Recovery occurred after approx. 5 minutes. Similarly, Zwick et al (6) found decrements in both the high and low contrast visual acuity of trained

rhesus monkeys within the first 2 to 3 min following exposure of the foveal area to a 532 nm Q-switched pulse. The laser had a repetitive pulse rate of 10 to 20 Hz, and produced minimal (50 μ) foveal lesions. They reported acuity recovered in 5 to 15 minutes following exposure. Merigan et al(7) showed that destruction of the fovea resulted in the loss of fine acuity at high luminance levels in the rhesus monkey. At lower target luminosity and for larger targets, no decrease in performance was noted. In a series of experiments designed to determine the effects of flashblinding stimuli upon the ability of both humans and rhesus monkeys to maintain compensatory tracking, Callin et al (8) reported that for the stimulus conditions (100 msec tungsten halogen flash and 20.7 μJ/flash), the average recovery time for each species was approx. 3 sec. A second study by this group using green or white (multiwavelength) laser pulses found no consistent effect upon tracking performance. The average flash recovery in those animals showing some disruption was approximately 2 sec. This was attributed solely to startle responses of the animals. Another interpretation of these data is that the fast recovery times exhibited by their trained animals was the result of the animal's ability to use parafoveal cues in tracking. This thus negated the central field flash effect. One method of determining foveal flash effects is to measure indirectly the integrity of the central retinal area by evaluating the cortical response to a pattern visual stimulus before and after a foveal laser exposure.

The pattern visual evoked potential (VEP) is an electrical response to a shifting stimulus composed of alternating light and dark bars recorded at the cortex. This potential primarily reflects activity in the fovea and the immediately surrounding macular area while suppressing perimacular involvement by insuring constant retinal illumination. Regan (9) has shown that the response of the electroencephalogram (EEG) to an alternating stimulus is one of entrainment of this signal at the alternating frequency. This phenomenon requires several seconds to appear following the onset of the stimulus. One hypothesis for this phenomenon is a neural recruitment of the retinal elements at the cortical level. The cortical elements then become synchronized to the signal.

Differences between human psychophysical data and corresponding electrophysiological results have been noted; recovery of the VEP is much faster than psychophysical recovery after response suppression by adaptation in a contrast threshold task (10). However, direct comparison of psychophysical and electrophysiological measures of contrast thresholds demonstrates a high correlation and indicates that the evoked potential can be an accurate reflection of perceptual experience (11 - 14).

The purpose of this study was twofold. First, determine if foveal flash effects could be identified and quantified by using an electrophysiological technique. Second, delineate those combinations

of variables, such as spot size and energy level, which would yield immediate and short-term changes in the visual system.

METHODS

Subjects: Nine eyes of seven cynomolgus (Macaca fascicularis) monkeys were used in the present study. The animals were sedated by intramuscular injection of ketamine HCl (10 mg/kg) and premedicated with atropine (0.008 mg/kg). An intravenous catheter was established to administer and maintain the dose level of the paralytic agent pancuronium bromide. The animal was intubated and breathing was maintained by a small animal respirator. The breathing and the electrocardiogram were monitored on a single channel of the physiological amplifiers throughout the experimental session. The eye of interest was dilated with 2% cyclopentolate HCl and 10% phenylephrine HCl. The animal was placed on an animal holder whose plane of rotation was adjusted to be in the center of the cornea of the experimental eye. The head was fixed and a lid speculum installed. Corneal clarity was maintained by frequent washes of normal saline (approx. every 10 sec). The unused eye was kept closed throughout the procedure. At the conclusion of the experiment, the paralysis was reversed with neostigmine and atropine.

Apparatus: Figure 1A is a diagram of the system used in this study. A Holobeam Series 300 Q-switched ruby laser operating at 694.3 nm with a pulse width of 20 nsec was coaxially aligned to the optics of a modified Zeiss fundus camera. Two spot sizes and two energy levels were chosen. The low dose level for minimal (50 μ: 0.2 degrees [°] visual angle) and large (500 μ: 2.0°) spot sizes were 18 and 178 μJ total interocular energy (TIE) as measured at the cornea respectively. At the higher dose level the TIE for the 50 and 500 μ retinal spot sizes were 39 and 422 μJ respectively. The fundus camera modification (Fig. 1B) consisted of a linear motion motor mounted on the side of the camera's optical system. The motor drove a high contrast square wave grating of either 1.6 or 2.8 cycles/degree visual angle in a square wave mode at 7 shifts/sec (3.5 Hz) in a focal plane conjunct with the retina. The normal field of view of the fundus camera is 30°. This was modified by the introduction of a field stop in the camera's final common path which reduced the projected grating to a stimulus diameter of 3.6° centered on the fovea. The VEP was recorded by a single subdermal needle electrode placed 1 cm superior to the inion and lateral to the midline referenced to linked ears. The signal was processed by a Grass 7P511 physiological amplifier. This signal was then recorded on FM magnetic tape while being analyzed on-line by a Nicolet MED-80 computer system and a PAR Vector Voltmeter. Off-line analysis was performed by playing back the taped signals into the MED-80 and/or Vector Voltmeter.

Procedure: The fovea was aligned with a reticle in the fundus camera field of view and the grating was focused onto the retina.

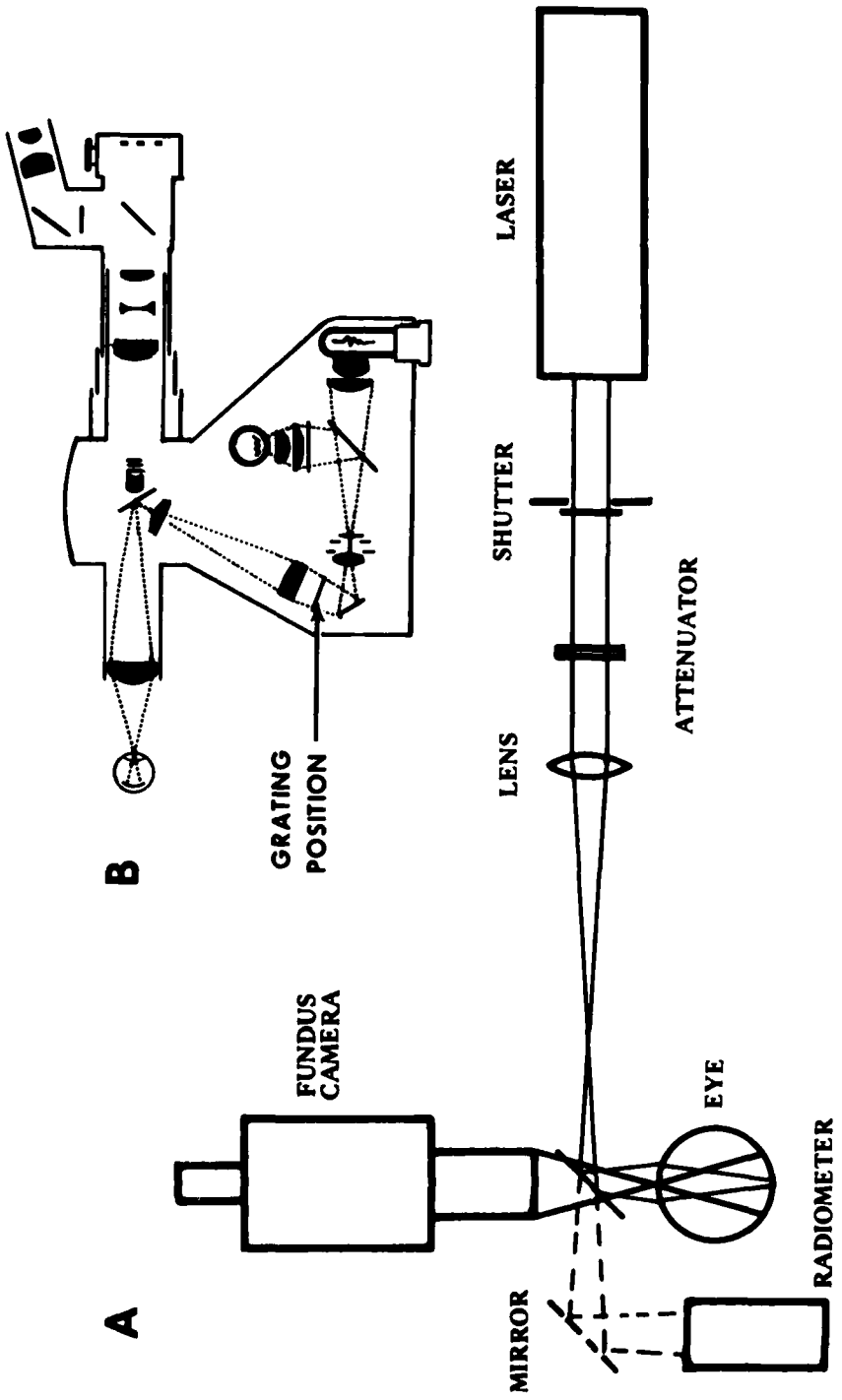

Figure 1. A. Laser and stimulus system arrangement. B. *(upper right)* Fundus camera diagram showing the position of the grating transparency.

Baseline VEPs were recorded in response to the oscillating grating. At this time, the aperture, if used, was introduced and further baseline data were obtained. During the stimulation, one or more single laser exposures was made to the fovea. Four measures of changes in the steady-state VEP were used, in addition to on-line observation of the averaged potential over short epochs. These measures were phase, magnitude, Pearson product moment correlation and the average standard deviation. Phase and magnitude traces were obtained by processing the VEP through the Vector Voltmeter synchronized with the 7 alteration/second grating stimulus. Changes in response phase reflect a change in the synchronization of the VEP and infer a loss of the ability of the visual system to follow the repetitive stimulus. The magnitude reflects the amplitude of the EEG component at the stimulation frequency. The Pearson correlation coefficient measure was obtained by comparing a pre-exposure averaged VEP (baseline) with sequential averaged VEPs (seven second epochs) recorded during the session. The correlation coefficient will theoretically approach 1.0 when the pre- and post-exposure VEP frequency elements show no difference in relative amplitude and phase. This measure is independent of absolute amplitude and ignores DC shifts. The average standard deviation measure is a mean variability estimate in relative units of the VEP processed in 11 sec bins (7 seconds averaging and 4 sec analysis time).

RESULTS

In the present study, under all of the stimulus and laser combinations: grating size (1.6 and 2.8 cycles/degree), field angular subtense (30^{o} and 3.6^{o}), high and low energy with large and small spot sizes, no immediate change in the VEP (i.e. within the first 5 sec) were observed. Neither were any long-term effects noted for those conditions in which minimal spot, low or high energy flashes were combined with large stimulus field sizes (5 eyes). However, marked changes occurred in the VEP as the post-flash interval increased for those conditions in which the high energy, large spot size and/or small stimulating field was used (4 eyes).

Data are shown in Fig. 2 for four animals under four different sets of conditions. The first trace shows phase changes in the VEP of monkey B2. A 30^{o} stimulus field produced a relatively stable phase locked response. The animal received a foveal exposure of 500 μ at 422 μJ, TIE. Little change in phase (digitized at 200 msec per point) was observed immediately after exposure. A later shift of 200^{o} was noted which began approx. 45 sec after exposure and continued for 15 sec. The second trace illustrates the response of monkey D3 to a Q-switched laser flash of low intensity (18 μJ) and minimal spot size (50 μ) with a test field of 3.6^{o}. A large 90^{o} phase shift occurred 90 sec. after the exposure. When the spot size of the laser exposure was increased to 500 μ (monkey B1) the 178 μJ TIF flash produced a large sharp phase shift of approx. 180^{o}, 106 sec after the flash.

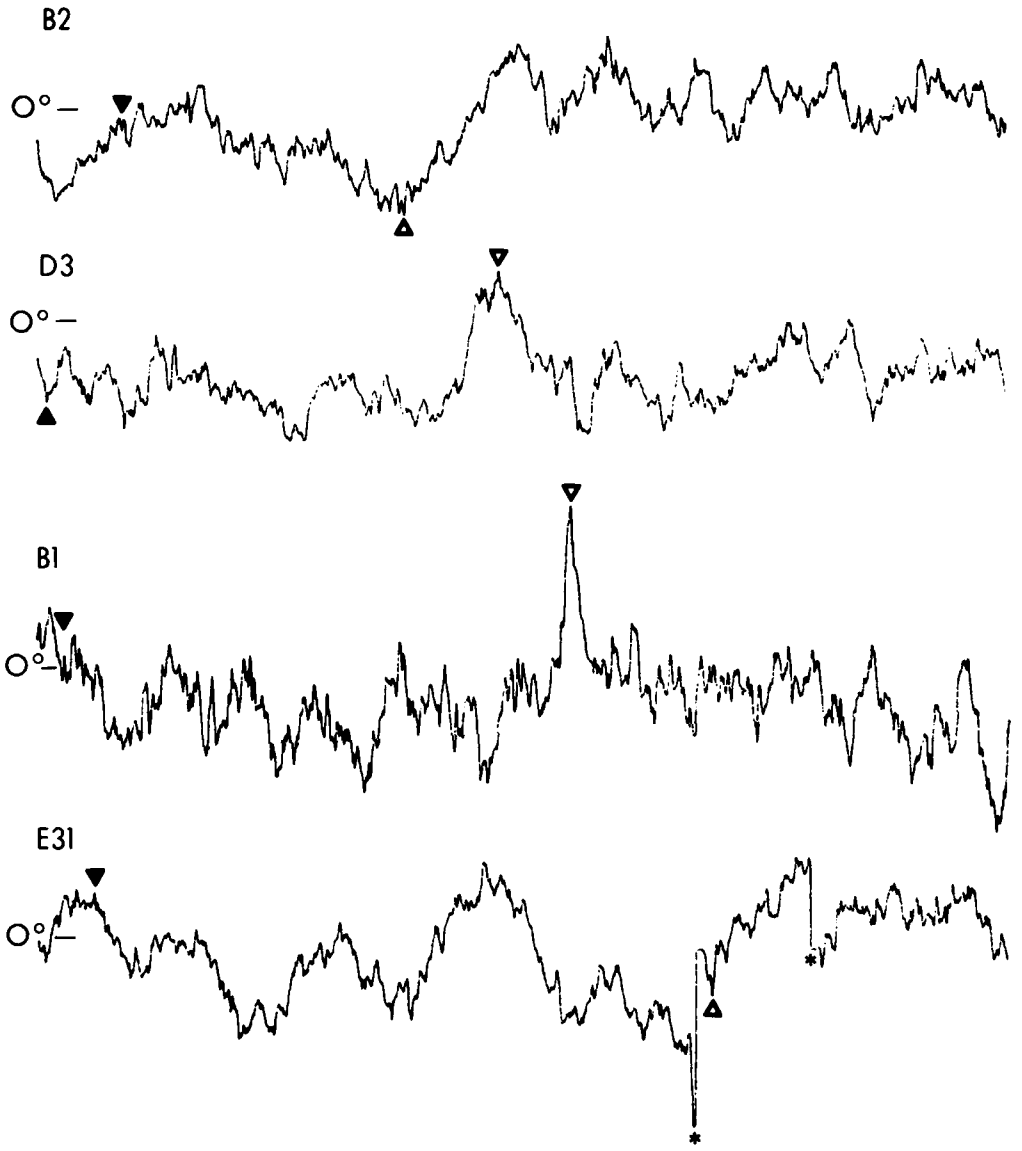

Figure 2. Phase recordings from the vector voltmeter for 4 animals. Dark triangles indicate laser exposure; light triangles indicate response. Asterisks on trace E31 indicate manual shift into and out of the neighboring phase quadrant. Total time of traces is 204 sec. See text for description.

The VEP quickly became resynchronized in this monkey and no further changes were observed. An increase in the laser energy to 422 μJ for the 500 μ spot size, 3.6° stimulus field condition produced a very large (approx. 250°) phase shift, 130 sec after the exposure (monkey E31). The asterisks in this trace mark the manual repositioning of the trace.

In addition to phase, three other measures of the changes in the VEP were recorded. These are shown in Fig. 3 for monkey D3. The first trace is a measure of the relative magnitude of the stimulus locked component of the VEP recorded simultaneously with the phase, phi (amplified from D3, Fig. 2). The point at which the magnitude approaches zero corresponds to the maximum of the phase shift. Line 3 shows the Pearson correlation coefficients for this epoch of data. A large decorrelation can be noted at the same time as the phase and magnitude shifts. The line marked sigma in Fig. 3 represents the variability of the VEP expressed as the mean of the standard deviations of the time-locked VEP 7-sec bin. The increased variability coincided with the shifts in magnitude, phase and correlation indices.

DISCUSSION

In the present study, little or no immediate flash effects were seen under any of the conditions used in this experiment. We have assumed that the VEP represents an ongoing, entrained response of the visual system to foveal events. Since we produced a visible change in the fovea with a Q-switched pulse, we would have expected to observe an immediate change in the VEP. This observation plus the fact that we observed a delayed change in the VEP leads to several possibilities. First, as Callin (8) has pointed out in his three studies of compensatory tracking performance, the flashes of laser light produced a momentary (2-3 sec) startle effect followed by a return to normal tracking behavior. In the curarized animal, this startle response would be absent. In Callin's (8) experiments, the animal's total tracking time was only 45 sec whereas in the present study, the effects did not appear until after 45 sec had passed. In addition, the data in the present study, as shown in Figs. 2 and 3, were recorded with time constants of sufficient length so as to mask a short 2-3 sec transient event.

Second, Zwick et al (6) showed data in which retinal lesions were produced by flashes resulting in visual acuity loss for considerable periods of time. Their immediate post-exposure measures were the average of the first 2-3 min post flash acuity data and thus do not reflect the nearer term continuous monitoring of the retinocortical response system. The perturbation reported in this paper may thus reflect the neurologic beginning of the phenomenon which was observed as an acuity decrement.

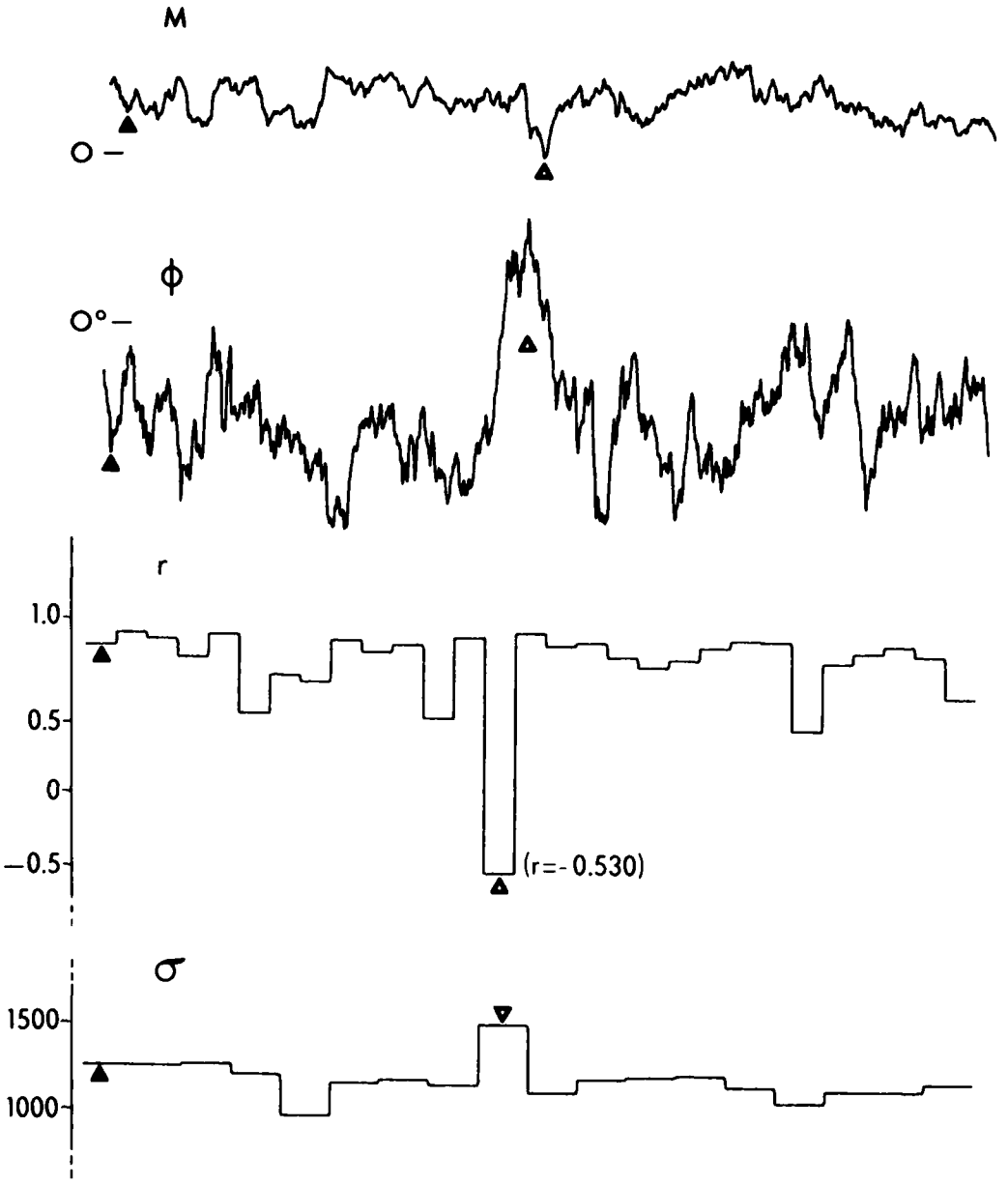

Figure 3. Analysis of animal D3 VEP. Magnitude (M), phase (phi), Pearson correlation coefficient (r), and average standard deviation (sigma) are shown. Dark triangles indicate laser exposure; light triangles indicate response. Total time of trace is 204 sec. See text for description.

Third, the short Q-switched pulse may not have been sufficiently long to produce a measurable flash response. In the earlier studies (5,6,8), longer (100 msec) single pulses were placed in the fovea. In another study (7) the Q-switched 20 nsec pulses were delivered in a 120 msec train at a rate of 10 to 20 Hz. While a VEP response to the flash itself may have been present, it may not have been of sufficient duration or persistence to be detected.

A fourth consideration is the effect of the wavelength of the flash upon the response. The Q-switched ruby laser pulse produces a 694.3 nm pulse which is near the spectral perceptual limit of the primate visual system. Randolph (15) has shown that flash blindness production was far inferior for a red (620 nm) source than for blue, green or white light flashes of equivalent energy. The studies previously cited used visible wavelength flashes closer to the peak of visual sensitivity. Thus the wavelength of the flash source may have contributed significantly to the lack of any immediate response.

Fifth, while the higher energy laser flashes at both the 50 and 500 μ spot sizes produced visible alteration at the exposure site, the changes observed may have been limited to the non-visual cell layer, the retinal pigment epithelium, with little accompanying effect upon vision as measured by the VEP.

While all of the aforementioned factors may have contributed to the finding of no immediate VEP response differences, the nature of the delayed response is such as to suggest a dual mechanism. Initially, edema develops at the site of the laser injury and extends laterally. The delayed effects noted in this study may reflect the disentrainment or desynchronization of the cortical response due to the mechanical displacement of the retina by edema. The subsequent recovery of the response indices may reflect the re-entrainment of the cortical elements due to recruitment among surviving retinal elements and may be independent of possible changes in visual acuity i.e. a non-visual phenomenon.

CONCLUSIONS

The 20 nsec Q-switched ruby laser exposures centered on the fovea produced no immediate changes in the grating visual evoked potential for 50 and 500 μ spot sizes at two energy levels even when visible changes occurred at these sites.

The findings may have been the result of the ruby laser wavelength, which was near the visual sensitivity limit of the eye, or of the single Q-switched pulse which may have occurred too quickly to produce an immediate and/or sustained change in the cortical response.

The observed delayed effects and subsequent apparent recovery of the VEP may reflect the development of edema at the laser exposure

site resulting in the desynchronization of the response for a period of time. This is followed by apparent recovery which was interpreted as recruitment of the spared retinal elements with subsequent neural re-entrainment at the cortical level.

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LASER OCULAR FLASH EFFECTS

Harry Zwick, PhD, Kenneth R. Bloom, BA,
David J. Lund, BS, and Edwin S. Beatrice, MD, COL MC

Recent developments of present ground laser systems are sufficient to pose a significant threat to complex man-machine interfaces. The viability of such interfaces is a vital component of the modern integrated battlefield. As a laser is a highly collimated source of light, under field conditions, visible or near infrared laser light will be focused to a very small spot on the back of the eye, the retina, where the sensory process for human vision is first initiated. Small burns from such minimal spot laser exposures can produce large changes in the immediate ability to detect critical detail under varying conditions. The extent of the impairment is unknown but long recognized as a critical aspect in evaluating the battlefield threat from low power laser systems. The present experiment was designed to determine if small spot laser flash exposure could produce transient changes in both high contrast visual acuity and resolutions of various acuity targets presented under liminal contrast conditions-contrast sensitivity. The data presented here indicate that repetitively pulsed Q-switched visible laser sources can produce transient changes in both high contrast acuity and contrast acuity and contrast sensitivity. These effects do suggest that complex target acquisition functions may be disrupted by small spot laser flash exposures that are likely from presently fielded Army laser systems.

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Biography of First Author

Present Assignment: Research Psychologist, Letterman Army Institute of Research.

Past Experience: Research Psychologist, Naval Air Development Center, Johnsville, Pennsylvania, 1968-1970,; Research Psychologist, Joint Laser Safety Team, Frankford Arsenal, Philadephia, Pennsylvania, 1970-1974; Visiting Scientist, Lawrence Berkeley Laboratories, Berkeley, California, 1977 to present.

Degrees Held: Bachelor of Arts, Earlham College, Richmond, Indiana, 1960; Master of Arts, Columbia University, New York, New York, 1963; Ph.D., University of Delaware, 1968.

Zwick, Bloom, Lund, Beatrice

LASER OCULAR FLASH EFFECTS

During this decade, lasers on the modern battlefield will become a directed energy threat to the eyes of ground force military personnel (1-3). One needs only to reflect on the enormous increase in electro-optical battlefield devices presently being developed to both train and equip troops for combat to suspect that a dramatic increase in accidental and intentional exposure incidence may well occur. Laser rangefinders (single pulse) and designators (multiple pulse) are anticipated to be commonplace in the modern electronic battlefield. While the future may hold to the concept of a laser injury as radiation that "vaporizes" its target, now we need only to be concerned with those devices that disrupt the complex man-machine interface by ocular injury. Such interfaces are critical to a modern equipped Army, and ocular injury will severely affect this complexity. Laser devices that inflict such ocular damage are easily available and will be prolific in ground battlefield scenarios. The present investigation was designed to incorporate several key features of the military scenario in order to address the question of a low level laser threat to the eye and acute vision.

To understand the nature of the low level laser threat, we need to understand some critical aspects of ocular anatomy, vision, and the nature of a laser exposure made under field conditions. The retina is the sensory tissue that is responsible for transducing light into simple visual sensation. Such sensations, processed as electrical impulses both within the retina and in the visual brain centers, produce our most complex visual experience. If a portion of this tissue is destroyed, vision can be permanently or temporarily altered. The photoreceptors, the actual biological transducers of light impulses into electrical impulses, are not uniformly distributed. The cone photoreceptors, concentrated in the fovea, are responsible for acute vision, maximal visual acuity, and fine spatial vision. If the fovea is damaged, visual acuity is dramatically altered and the visual scene is blurred. The fovea, a relatively small piece of the human sensory retina, is at the center of the optical axis of the eye. Thus, use of military optics places the fovea in a most vulnerable position.

In several previous experiments we demonstrated that foveal laser exposure could dramatically alter the ability to resolve fine spatial detail, i.e. visual acuity (4,5). These experiments demonstrated both permanent, as well as transient changes in acuity associated with

moderate to low levels of laser flash exposure. But all of these previous experiments were done with relatively large retinal spot sizes (150 to 350 μm). In the field, however, the laser flash will produce a small retinal spot (30 to 50 μm) because of the laser's low divergence. Our questions in the present experiment were: will such exposure produce a significant effect on the retina and visual acuity? Will small punctate foveal exposures have an effect on visual acuity similar to that of larger retinal spot exposures, where the entire fovea was involved? Will small spot foveal exposure affect retinal areas outside the fovea?

By measuring contrast sensitivity across a range of acuity target sizes, we were able to assess the possible lateral influence of small spot laser flash exposure. While visual acuity expresses the very finest target that can be resolved spatially, it does not reflect spatial visual function for larger targets. In order to measure the effects on liminal spatial vision for large as well as small targets we measured contrast thresholds as well as visual acuity. The reciprocal of the minimum contrast ratio threshold is contrast sensitivity, and one may plot a function relating contrast sensitivity to acuity target size or spatial frequency. In the contrast sensitivity function the smallest resolvable target has the lowest contrast sensitivity, requiring the highest contrast threshold.

In this experiment we have not only simulated battlefield exposures with small retinal spot exposure but, because many field laser systems are repetitively pulsed, we have also employed a repetitively pulsed laser system. The wavelength of the laser source (532 nm) is close to the peak of the daytime maximum color sensitivity of the human and monkey eye (550 nm).

METHODS

Rhesus monkeys were trained on a visual acuity task in which exposure to a laser flash could be administered during task performance (4-6). Animals were trained for many months to discriminate bright achromatic Landolt ring acuity targets, rings with gaps ("C"s), from other bright ring targets that lack this gap ("O"s). The minimum resolvable spatial detail (visual angle) for the rhesus monkey is similar to that of the human.

The rhesus is also quite comparable to the human in its minimum contrast threshold for various target sizes. To measure contrast sensitivity, the gap of the Landolt ring was expressed as an aperiodic spatial frequency.

In order to expose an animal and track the immediate visual consequence of a laser exposure in a reliable manner for at least a half hour post-exposure, very stable visual function baselines were

required. Spatial vision thresholds (visual acuity and contrast sensitivity) were determined by a method that allows instantaneous determination of threshold (7). The rhesus were trained to titrate either the size or the contrast of the acuity targets about their threshold. Following initial discrimination learning and determination of acuity or contrast sensitivity for a given acuity target, animals were trained to yield highly stable baselines with minimum variation over a period of at least an hour. Stability criteria of approximately 0.2 to 0.4 log units in either acuity or contrast sensitivity maintained over a 30 to 60-minute period were generally required before any animal was considered ready for exposure.

For two animals trained to track only their high contrast acuity, three to four exposure sessions were given to establish reliability of a given exposure level. Exposure levels were increased until a criterion deficit of about 90% of baseline acuity could be obtained for about 2 minutes post-exposure. For three contrast sensitivity animals, the effect of laser flash exposure was determined on one spatial frequency at a time for three to four exposure sessions.

Laser flash effects were obtained for small spot pulsed visible laser (532 nm) flash exposure on visual acuity, and contrast thresholds for spatial frequencies from 38.5 cycles/degree to 2.2 cycles/degree, (i.e. Snellen acuity notation from 20/15 to 20/267). All exposures were made with the flash presented through the gap in a 0.78 minute of arc (20/15) Landolt ring. This assured foveal exposures, as the fovea was required for accurate discrimination of this size target. An exposure was made if the daily pre-exposure baseline was within 0.4 log units of the previous session's baseline for acuity or contrast sensitivity. A frequency doubled neodymium laser (532 nm) operating at 20 Hz was used. Exposure consisted of six 20 nsec pulses delivered within a 300 msec time window. In the early portions of this experiment a 10 Hz pulse repetition frequency (PRF) was employed and six pulses were cumulated over three successive trials spanning a period of 36 sec. The average calculated nominal total Intraocular energy (TIE) per pulse was 3.0 μJoules for a 5 mm pupil in high contrast acuity recovery tests. All contrast threshold functions were measured against a 70 ft-Lamberts background and, therefore, a 3 mm pupil was assumed for calculation of the total intraocular energy (1.1 μJoules). All TIE levels are nominal values with variation of TIE about $\pm$20% within a given animal.

Five animals were used in these experiments. All had pretraining refractive errors of less than a 1/2 diopter; all had normal appearing fundi prior to exposure. Fundi of selected animals were reexamined only after an entire exposure series for the given animal was complete.

RESULTS

An acuity threshold session, lasting about 60 minutes, from one animal is presented in Figure 1. Landolt rings presented to this animal immediately after exposure had to be increased in size to a Snellen acuity level of 20/108 before tracking of post-exposure recovery could be achieved. This post-exposure threshold acuity level

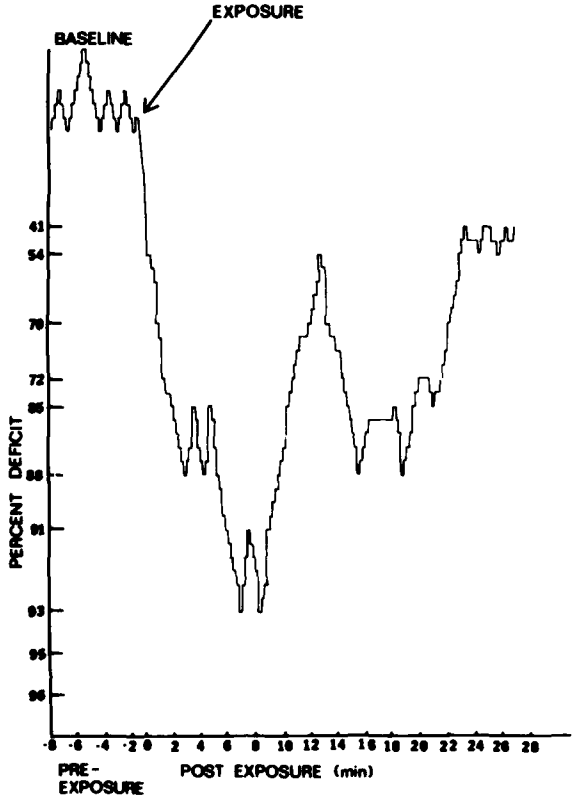

Figure 1. Raw data taken from an acuity exposure session is shown. The Y-axis is an ordinal scale where the location of Landolt rings in a size graded series of acuity targets is expressed as a percent deficit relative to the pre-exposure baseline. The last 8 minutes of the pre-exposure acuity baseline is followed by a laser exposure. A maximum acuity deficit of approximately 93% was seen as long as 8 minutes post-exposure. At 28 minutes, recovery was to about 50%, with a full recovery occurring within 1 hour post-exposure.

corresponded to at least a 90% deficit with respect to pre-exposure acuity baseline, and was persistent for several minutes post-exposure. Even after recovery began, it was slow and incomplete at the end of 30 minutes. At the end of the actual 50 minute post- exposure session, however, the animal's acuity baseline had returned to pre-exposure levels. No subsequent deficit was noted on successive test sessions.

In Figure 2, recovery curves for visual acuity (2a) and contrast sensitivity (2b) are presented. Two acuity recovery functions are shown in 2a. Both of these functions were obtained in a manner similar to that shown in Figure 1, except here we have averaged the acuity for each two minute block of post-exposure data over several exposure sessions for one animal. The lower curve was obtained from exposure to 6 pulses at a 10 Hz pulse repetition frequency, while the upper curve was obtained from exposure to 6 pulses at a 20 Hz pulse repetition frequency. In the former, six pulses were delivered in 2

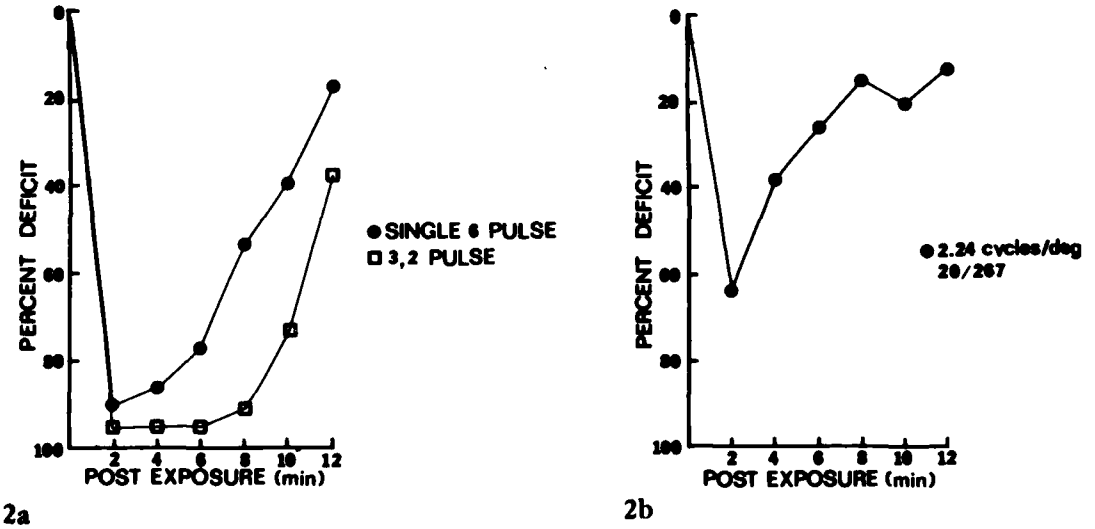

Figure 2. Post-exposure recovery curves for acuity (2a) and contrast sensitivity (2b) are presented as percent deficit relative to pre-exposure baselines. Significant initial deficits and recovery from such deficits are represented. The recovery curve for contrast sensitivity (2b) was measured with a large target having a spatial frequency of 2.24 cycles/degree. The acuity recovery curves represent the average of 4 exposure sessions for each condition, while the contrast sensitivity recovery curve is an average of 20 exposure sessions across 3 animals. By inspection, these average curves are highly representative of all individual recovery functions.

pulse bursts on three consecutive Landolt ring trials over a 36-sec period, whereas in the latter curve the 6 pulses were delivered in a 300 msec interval on a single Landolt ring trial. While a more prolonged recovery time is evident for the 10 Hz repeated trial exposure, both require close to 20 minutes for full recovery to occur.

Figure 2b is a recovery function of the contrast sensitivity for an acuity target equivalent to 20/267 Snellen, or a spatial frequency of 2.2 cycles/degree. It is a large target that involves foveal as well as parafoveal stimulation. Initial deficits in contrast sensitivity, and recovery times for such size targets were essentially the same as those for targets requiring much smaller foveal areas. Similar curves for 20/15 Snellen acuity targets, or spatial frequencies of 38.5 cycles/degree, were essentially equivalent in the time course of recovery. Both small and large targets showed little recovery during the first two to four minutes post-exposure. After the initial four minutes post-exposure, full recovery required about 15 minutes. Post-exposure contrast threshold 95% confidence limits did not overlap those of baseline in any specific animal until about 6 minutes post-exposure. Statistical significance of post-exposure relative to pre-exposure thresholds for $p<.05$ was obtained during the first 4 to 6 minutes in all animals. Recovery became more variable both within and across animals beyond 6 minutes post-exposure.

Although an immediate and substantial deficit in spatial vision was usually produced, not every exposure produced the same initial loss. The histograms of Figure 3 show the percentages of exposure trials required to produce criterion deficits of 40, 60, or 70 % within the first 2 min post-exposure. Data were taken from three contrast sensitivity animals from all exposures where 38.5, 10.0, and 2.24 cycles/degree were used as test spatial frequencies. A total of 92 exposure sessions are represented.

Most exposure sessions produced a 40% deficit over the first 2 min of exposure regardless of spatial frequency. About 90% of all exposures tabulated produced a deficit >40% for these initial 2 min post-exposure; about 60% of all exposures produced deficits >60%; and, between 35 and 60% produced deficits >70%. In all three categories, the largest spatial frequency was affected at least to the same degree as was the finest spatial frequency. The intermediate spatial frequency seemed somewhat less affected than those at the spatial frequency extremes. When the complete spatial frequency spectrum is plotted, it is evident that intermediate spatial frequencies were somewhat less affected initially and recovered more rapidly than either the largest or the finest spatial frequencies.

corresponded to at least a 90% deficit with respect to pre-exposure acuity baseline, and was persistent for several minutes post-exposure. Even after recovery began, it was slow and incomplete at the end of 30 minutes. At the end of the actual 50 minute post- exposure session, however, the animal's acuity baseline had returned to pre-exposure levels. No subsequent deficit was noted on successive test sessions.

In Figure 2, recovery curves for visual acuity (2a) and contrast sensitivity (2b) are presented. Two acuity recovery functions are shown in 2a. Both of these functions were obtained in a manner similar to that shown in Figure 1, except here we have averaged the acuity for each two minute block of post-exposure data over several exposure sessions for one animal. The lower curve was obtained from exposure to 6 pulses at a 10 Hz pulse repetition frequency, while the upper curve was obtained from exposure to 6 pulses at a 20 Hz pulse repetition frequency. In the former, six pulses were delivered in 2

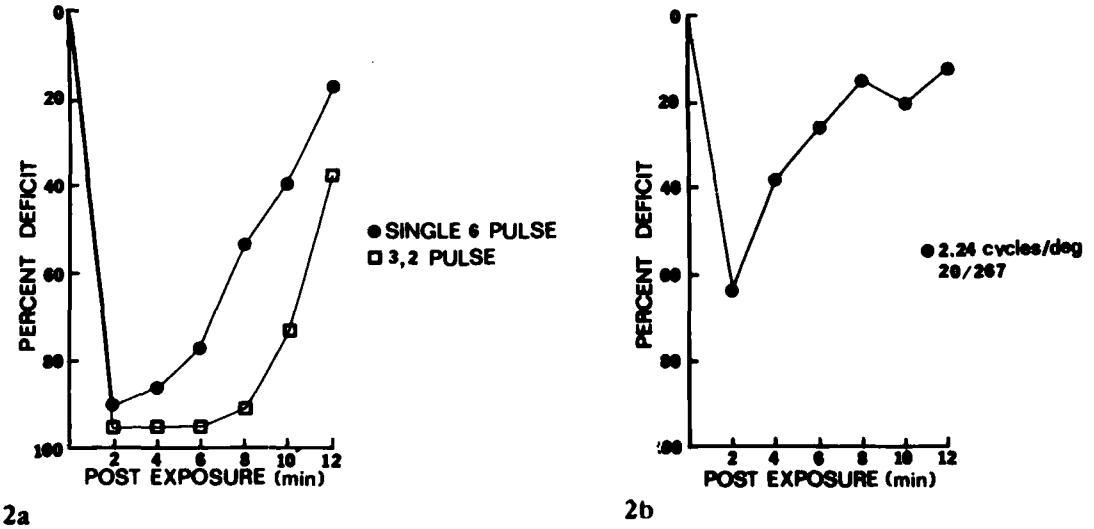

Figure 2. Post-exposure recovery curves for acuity (2a) and contrast sensitivity (2b) are presented as percent deficit relative to pre-exposure baselines. Significant initial deficits and recovery from such deficits are represented. The recovery curve for contrast sensitivity (2b) was measured with a large target having a spatial frequency of 2.24 cycles/degree. The acuity recovery curves represent the average of 4 exposure sessions for each condition, while the contrast sensitivity recovery curve is an average of 20 exposure sessions across 3 animals. By inspection, these average curves are highly representative of all individual recovery functions.

pulse bursts on three consecutive Landolt ring trials over a 36-sec period, whereas in the latter curve the 6 pulses were delivered in a 300 msec interval on a single Landolt ring trial. While a more prolonged recovery time is evident for the 10 Hz repeated trial exposure, both require close to 20 minutes for full recovery to occur.

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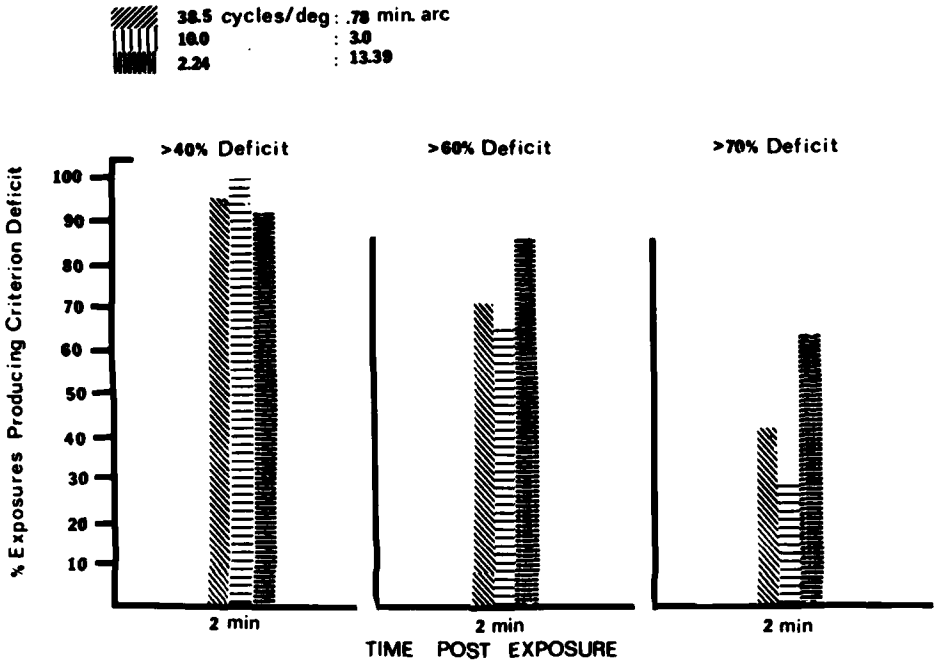

Figure 3. The degree of maximum deficit in contrast sensitivity was not uniform for all exposures. Variability can be seen in the amount of post-exposure change, with less than 50% of the exposures resulting in deficits greater than 70% of baseline levels, while deficits greater than 40% were produced on more than 90% of all exposures. Deficits greater than 60% were produced on 60-80% of the exposures. These data represent a total of 92 exposure sessions across 3 animals.

Several exposure sessions produced deficits in either acuity or contrast sensitivity that lasted more than the duration of the test session, and appeared more selective to the highest spatial frequencies. Such effects, however, were difficult to quantify fully because of their infrequence in the present study. However, recovery from these exposures always occurred with several post-exposure sessions.

Fundus observations of animals examined after the completion of all laser exposure sessions revealed small punctate lesions in the foveal areas including the foveola, the central portion of the fovea. A fundus photograph taken from one of the subjects is shown in Figure 4.

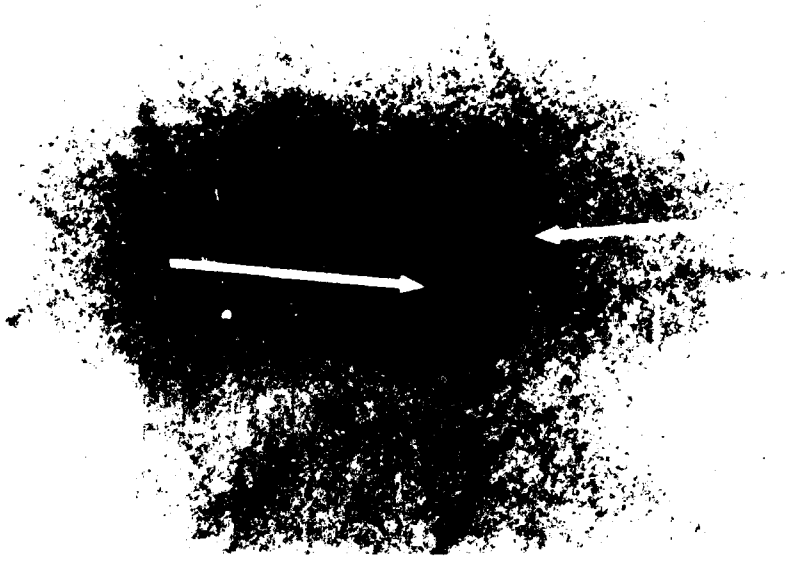

Figure 4. Rhesus fundus showing "punctate" foveal lesions. Arrows indicate the location of the lesions.

DISCUSSION

We have demonstrated that small spot laser flash exposures produce transient changes in high contrast visual acuity and contrast sensitivity. These data indicate that effects may involve areas greater than the predicted retinal image diameter of 30 to 50 μm.

Several possible explanations are applicable to these findings. Most exposures in this experiment involved trains of Q-switched pulses. Such exposure conditions in combination with small eye movements may have "painted " a larger effective retinal spot across the fovea than would have been possible with a single pulse alone. In our early experiments, we used a 10 Hz PRF, and distributed six pulses over a 36-sec period, 2 pulses per trial. This exposure condition (Figure 2a-lower curve) produced a longer lasting initial deficit than a single burst of six pulses delivered over 300 msec.

As most exposures made were at the ED_{50} for retinal burn criterion, a second factor, local retinal edema, is possible as well. Local edema and its spread to neighboring retinal areas might well have affected parafoveal areas for a short period of time. Such a phenomenon was suggested to explain analogous effects for total foveal damage (4).

Neural retinal interactions are a third element in the explanation of these effects. The visual fields of foveal photoreceptor neural

systems (foveal receptive fields) typically are considered to be small, subserving a small number of photoreceptors located within the fovea. While these results do not comment on the size of foveal receptive fields directly, they suggest that foveal receptive fields may overlap parafoveal areas, so that a foveal alteration could affect parafoveal processing of photoreceptor input.

In addition to producing large changes in fine spatial vision, these flashes frequently produced initial delays in the recovery function. Such delays were generally evident during the first 2 to 4 minutes following flash exposure. A similar finding has been reported for longer pulse widths (100 msec) and larger retinal spot sizes (150 to 350 μm) (5). Such delays in recovery after flash increased with exposure power up to permanent acuity losses. Temporary delays in recovery may be reflective of foveal neural "inelasticity" or foveal "blanking" as well as local retinal edematous changes.

Permanent changes lasting more than a single session were not as evident as those obtained in previous work, where large spot-induced fovial damage produced long-term loss in visual acuity that required at least six months for recovery, and longer term residual loss in color vision (4,8). In the present experiment, while portions of the fovea may be damaged, the long-term effects are either non-existent or difficult to measure, perhaps requiring more sensitive spectral measurement(4,8). Nevertheless, obvious lesions were produced in and around the fovea. The possibilities that foveal receptive fields are larger or more dynamic than originally conceived (9,10) or normal foveal function can be maintained by the "spared" foveal areas are suggested as explanatory factors.

These results have significant implications for lasers on the modern battlefield. We have shown that minimal spot laser exposure can affect liminal spatial visual function, as well as foveal retinal tissue. Tactically, a compromise in the ability to resolve fine spatial detail in low contrast ground environments may produce an immediate field casualty. Subtle changes in lighting conditions as occur at dawn or dusk, in target reflections and glare, contribute to altering subtle contrast and fine spatial detail required in any complex target acquisition task. Under such conditions, a small induced change in minimal spatial resolution threshold or contrast threshold could result in failure to acquire a critical target. On the other hand , many military scenarios require acquisition of high contrast targets where the requirement for the resolution of fine spatial detail is less stringent. Such situations could be affected to a lesser degree by point source flashes(11), although alteration of retinal foveal tissue is still as likely as in the above military scenario.

We have shown that laser flash exposure can produce significant changes in fine spatial vision and that such effects can be produced

with small spot retinal flashes that are likely from highly collimated fielded laser systems. These effects may produce injury to the retina as well as transient loss in fine spatial detail and alteration of normal contrast requirements for optimal target acquisition functions. Such transient effects may, therefore, interfere with mission completion as well as having unknown long-term consequences for normal foveal vision. These effects represent a potentially serious Army field hazard regarding both present and near term development of Army laser systems.

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Schuschereba, Beatrice

AUTORADIOGRAPHY OF PRIMATE RETINA AFTER Q-SWITCHED RUBY LASER RADIATION

Steven T. Schuschereba, MA, and Edwin S. Beatrice, MD, COL MC

An adult Rhesus monkey was exposed to Q-switched ruby laser (Q-SRL 694.3nm) radiation above, at, and below threshold levels (ED_{50} = 202 µJ) to determine the effects on amino acid ([^{3}H]-L-leucine-4,5) incorporation in lesion sites 2.5 and 5 days after exposure. Light microscopy revealed that ophthalmoscopically sub-visible lesions showed alteration at the retinal pigment epithelium photoreceptor interface. Autoradiography revealed that lesions that were not clearly visible histologically showed altered metabolism. Rod outer segment renewal rates (ROSRRs) and outer nuclear layer grain counts (ONLGCs) for the 2.5 and 5-day-old lesions were reduced at 2.5 days and increased, at 5 days. ROSRRs in the 1000 µJ 2.5-day lesion showed more inhibition than in the 5-day lesion. However, ONLGCs were increased over normal, which suggest a blockade lesion. However, ONLGCs in the 5-day lesion were increased 153% over normal, which suggest a blockade of protein flow proximal to the inactivated protein synthesis compartments. Conversely, the 100 µJ lesions at 5 days showed the greatest increase above normal (+15%) in ROSRRs with comparable increased ONLGCs (+18%) suggesting an overall increase in protein synthesis. Furthermore, cones in the center of the 756 µ lesion showed a selective absence of label, which suggest that cones are more sensitive than rods to Q-SRL radiation. Some cones in the 5-day macular lesion showed outer segment membranes in the inner segment. These have previously not been described for laser-exposed retinas. These changes may represent permanent alterations in photoreceptor metabolic pathways and, in turn, perhaps permanent decrements in visual function. As a result, ocular safety in the use of laser devices must involve the critical evaluation of the currently used ED_{50} and maximum permissible exposure (MPE) levels which are derived from these levels (by factors of 10 to 100) and the long-term considerations of these findings.

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Biography of First Author

Present Assignment: Electron Microscopist, Letterman Army Institute of Research, Presidio of San Francisco, CA

Past Experience: Biological Assistant, Joint AMRDC-AMC Laser Safety Team, Frankford Arsenal 1973-1974. Electron Microscopist, Letterman Army Institute of Research, Presidio of San Francisco, CA, 1975 to present.

Degrees Held: Bachelor of Science, Cornell University, Ithaca, NY, 1972. Master of Arts, San Francisco State University, San Francisco, CA, 1982

Schuschereba, Beatrice

AUTORADIOGRAPHY OF PRIMATE RETINA AFTER Q-SWITCHED RUBY LASER RADIATION

Biological implications in the use of laser devices by the Army has been the subject of study in the last two decades. For laser systems operating in the visible and near-infrared spectral region, the retina is the most vulnerable part of the body, and retinal effects have been extensively characterized ophthalmoscopically, histologically, and functionally (1-7). These effects vary from severe damage, accompanied by massive retinal hemorrhaging and extrusion into the vitreous (6), to small retinal burns characterized by focal retinal opacities (5) to changes in visual function (7) with or without morphologic changes.

A principal concern of laser bioeffects investigations has been mechanism and recovery processes of retinal tissue after damage by laser radiation near the visible damage threshold. Adams et al (2), in their study of threshold exposure limits, found that extremely low output energies from a Q-switched ruby laser (70 uJ at the cornea) produced extensive changes in the rhesus monkey retina when examined by electron microscopy. These changes included formation of bizarre fingerprint-like whorls in the outer segments at the base of the outer segment in the photoreceptor cells. This is in sharp contrast to the highly ordered arrangement in normal photoreceptors. The low levels of coherent radiation were thought to affect the formation of the outer segment membranes. Similar findings are observed in aged human retinas (8).

Young (9) studied outer segment formation by autoradiography, a technique that used radioactive amino acids as precursors for proteins, and discovered that an enormous amount of metabolic effort is devoted to a process that supports visual function. Rods, which subserve scotopic vision (vision in dim light), and cones, which subserve photopic vision (vision in bright light), expend most of their energy on the repeated replacement of their photon-capturing devices, the outer segments. Proteins were synthesized in the inner segment and migrated into the outer segment where they were assembled into membranous discs that comprised the outer segment stack. Opsin, the rod visual pigment protein, comprised most of the protein in rod outer segments. Smaller quantities of proteins are produced in the nucleus and very little in other parts of the cell. The question of whether or not near threshold laser light exposure can alter photoreceptor metabolism has long been pondered. Marshall (1) first reported, for suprathreshold Q-switched ruby laser exposures of the

retina, that protein synthesis (in vitro) was inhibited in photoreceptor inner segments and nuclei; however, no work has been reported on the effects of laser light on outer segment synthesis rates. It seems that knowledge of how lasers alter normal biologic function and cellular dynamics is an important area of study that should parallel other studies of ophthalmoscopic, histologic, and functional changes. The study of photoreceptor metabolism after Q-switched ruby laser radiation might give additional information regarding damage mechanisms involving metabolic activities. An understanding of the metabolism of damaged retinal tissue might suggest methods of treatment for such injury. In addition, such an understanding may also pave the way for preventive measures against injury. To gain insight into how lasers affect photoreceptor metabolism a study was undertaken to examine laser-induced retinal changes in outer segment renewal rates by autoradiography.

The work objectives of this study were: 1) to evaluate the recovery of Rhesus monkey retinal rods (similar in retinal structure to the human) after exposure (20 nsec pulse duration, 1000 μm beam spot-size) to a range of Q-switched ruby laser radiation (suprathreshold to subthreshold for retinal damage as evidenced by ophthalmoscopy), 2) to correlate morphologic damage by both light and electron microscopy with biochemical abnormalities observed by autoradiography, 3) to assess the degree of sensitivity provided by autoradiography and 4) to postulate possible damage and recovery processes.

MATERIALS AND METHODS

The Q-switched ruby laser system was employed (4). This system had been previously used to determine ED_{50} levels for 1000 μm diameter exposures (10) and transmission electron microscopy studies (2). The output beam diameter was 1.7 mm with beam divergence of one milliradian. A 20 diopter convex lens placed at twice the focal length of the lens (10 cm) produced a 2.2 mm beam on the cornea with a 67 mr divergence resulting in 1000 μm retinal irradiance diameter for a single 20 nsec pulse. An ultrafast detector and oscilloscope were used to measure each exposure. A helium-neon (He-Ne) laser of low output energy was aligned colinearly to aim the ruby beam on the retina at predetermined loci.

A Rhesus monkey was anesthetized with halothane. The pupils were dilated and corneal transparency was maintained by irrigation with normal saline. The retina was viewed and photographed through a Zeiss fundus camera immediately before and after laser exposure. An artifactual optical target marker in the fundus camera coaligned with the He-Ne laser was used to denote the exact lased area on fundus photographs.

Single 20 nsec laser exposures were made in the left fovea (756

μJ) and concentrically around the macula at 1000, 400, 200, and 100 μJ (Figure 1). Two and a half days lapsed and a similar exposure scheme was repeated with the exposure sites located in a circle 3 mm internal to the first. Within one hour of the last exposure, the animal was injected with 70 mCi of [^{3}H]-L-leucine-4,5 (specific activity, 30,000-60,000 mCi/mmol) in 2 ml of 2% ethanol via the catheterized right saphenous vein. Two and a half days after the ^{3}H-leucine injection, eyes were enucleated.

One square millimeter blocks (of lesions and controls) were dissected and fixed in Karnovsky's fixative (11). All tissue processing and autoradiography were performed by a previously described technique (9).

The total length of the rod outer segments and the distance from the outer-inner segment junction to the center of the transverse band of radioactivity on the outer segments in autoradiographs were measured by an ocular micrometer at X1000 (Figure 2). Rod outer segment renewal rates (ROSRRs) were determined by dividing the total outer segment length by the distance of the band of label from the outer-inner segment junction multiplied by the number of days after injection. Slides with lesions of maximum diameter for each exposure site and adjacent controls were selected. Measurements were taken at 50 μm intervals across given radii of the lesions from the established center. Measurements were in microns on five rods at each interval for each lesion and control.

Outer nuclear layer grain counts (ONLGCs) were tabulated and compared only within the same section (i.e. grain counts of the lesion center were compared with grain counts at the lesion edge). Counts were made on two different sections from two different slides for each lesion. The difference between the two areas was computed as a percent difference.

The retinal pigment epithelium (RPE) thickness was measured using a linear reticle at X1000. A minimum of five measurements, approximately 10 μm apart, were taken from the lesion center and periphery on three sections.

RESULTS

Little histopathologic difference existed between the 2.5 and 5-day lesions, with the exception that the 5-day lesions were smaller in size. In general, a decreasing severity in pathology occurred with decreasing exposure energy and with increasing distance from the lesion center. The most remarkable morphologic findings were that cones were more easily and more severely disrupted than rods and that cones in the macula showed outer segments in their inner segments. The remainder of the pathologic findings are summarized in Table 1.

Mean RPE thickness values for all the 2.5 and 5-day lesions showed

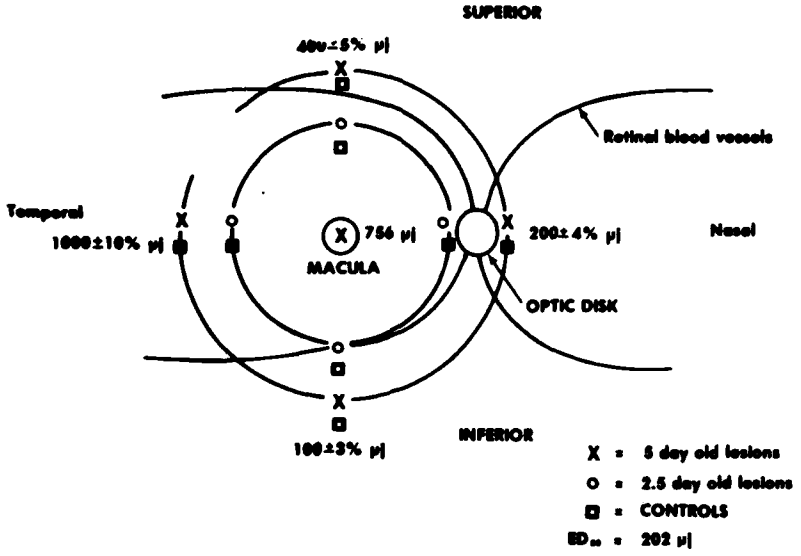

Figure 1. Fundus schematic of right-eye and laser exposure locations.

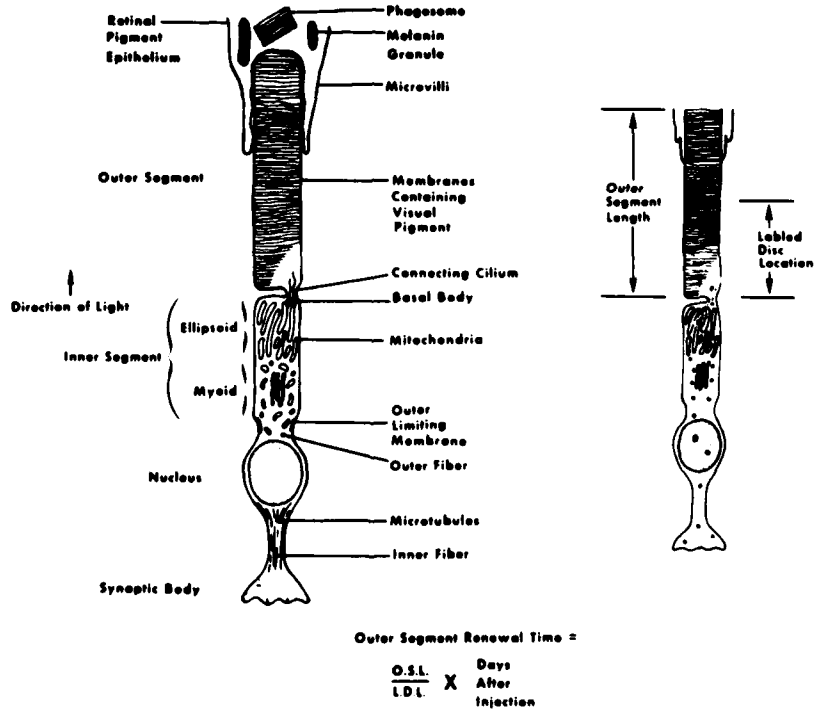

Figure 2. Diagram illustrating the components of a vertebrate visual cell. Formula for estimating rod outer segment renewal is at the bottom of the figure.

Table 1

Pathologic findings in a rhesus monkey retina (9 lesions, 5 sites*) after irradiation with a Q-switch ruby laser (694.3 nm)
Plus sign (+) indicates presence of characteristic; blank space indicates absence of characteristic

MORPHOLOGIC CHARACTERISTICS†	EXPOSURE ENERGY (μJ)								
	1000		756	400‡		200		100	
days after exposure	2.5	5	5	2.5	5	2.5	5	2.5	5
Hemorrhage (subretinal and preretinal)	+	+							
Loss of photoreceptors	+	+	+						
Vacuolization and depigmentation in choroid	+	+	+						
Melanin granule fracture	+	+							
Cellular proliferation of RPE	+	+	+			+	+		
Pyknotic photoreceptor nuclei	+	+	+						
Cone inner segment disruption	+	+				+			
Dissolution of outer segments	+	+	+						
Inclusion material in the outer receptor fibers			+						
Inner retina vacuoles	+	+	+						
Outer segments in the inner segments			+						
Melanin granule clumping	+	+	+	+	+	+	+	+	+
Pyknotic RPE nuclei	+	+	+	+	+	+	+		
Lamellar disarray of outer segments	+	+	+	+	+	+	+	+	+
Pyknotic outer segment debris						+	+		
Vacuolated inner segments	+	+	+	+	+	+	+	+	
Macrophages in subretinal space	+	+	+			+	+	+	
Hypopigmentation of RPE	+	+	+	+	+	+	+	+	+
Retraction of outer segments from the RPE	+	+	+	+	+	+	+		
Decreased length and thinning of outer segments	+	+	+	+	+	+	+	+	+
Reduced RPE thickness	+	+	+	+	+	+	+	+	+
Disorganized RPE microvilli	+	+	+	+	+	+	+	+	+

* Only one lesion was placed in the macula and analyzed 5 days later.
† Efforts have been made to list the characteristics by a decreasing order of severity.

‡ The distortion of the beam energy profile produced a distorted energy deposition and an under-represented response in the retina.

that the RPE thickness was reduced as the function of the exposure energy in the center of the lesion (Figure 3). An increase in RPE thickness occurred at the periphery of all the lesions. The range of RPE change in the macula was comparable to the values obtained for the 200 µJ extramacular lesions which suggests an attenuation effect to the RPE.

In contrast to the histologic findings, large autoradiographic variations occurred between the 2.5 and 5-day lesions. In the 1000 µJ 2.5 day lesion center (Figure 4), a decrease in label intensity was noted in the rod outer segment band as well as in the outer nuclear layer. The inner retina and RPE cells in the center of the 1000 µJ 5-day lesion showed a greater label uptake than peripheral or unexposed regions. With increasing distance from the edge of the 1000 µJ lesion to the center, cone inner segments contained less label (Figure 4).

Many undulating peaks occurred in the band of label in the 400 µJ lesions. These peaks suggested a distortion in the geometry of the laser beam energy distribution. In the 200 µJ lesions the label intensity in the rod outer segment band was decreased in the center of the lesion. In the 100 µJ lesions, the band of label was fragmented laterally.

All of the 2.5-day lesions showed less inhibition in ROSRRs with decreasing exposure energy (Figure 5). All the 5-day lesions showed more nearly normal ROSRRs (Figure 6). In either the 2.5 or 5-day 1000 µJ lesions, almost no rod outer segment renewal was taking place. Only the 5-day 1000 µJ lesion showed some rods with label in the outer segments. In the 5-day 200 and 100 µJ lesions, ROSRRs were increased 10-15% above the normal rate.

Calculated times for complete outer segment renewal are based on measurements taken on photoreceptors (Table 2A) and are given in Table 2B. Since outer segment lengths were slightly variable, depending upon the region of the retina where the outer segments were measured, estimated normal and abnormal renewal times varied accordingly. Rod outer segment renewal time projections are based on the development of events in the 2.5 and 5-day lesions and are expected to vary and develop further. After high energy laser exposure (1000 µJ), rods will take about three times as long to synthesize and renew their normal complement of outer segment discs. Laser exposures at 200 and 100 µJ (subvisible lesions) cause rods to synthesize and renew their normal complement of outer segment discs more rapidly (Table 2B).

ONLGCs from the centers of lesions were compared with values from the lesion periphery (600 µm from the lesion center) (Figure 7). ONLGCs for the 2.5-day 1000 µJ lesion centers showed a marked

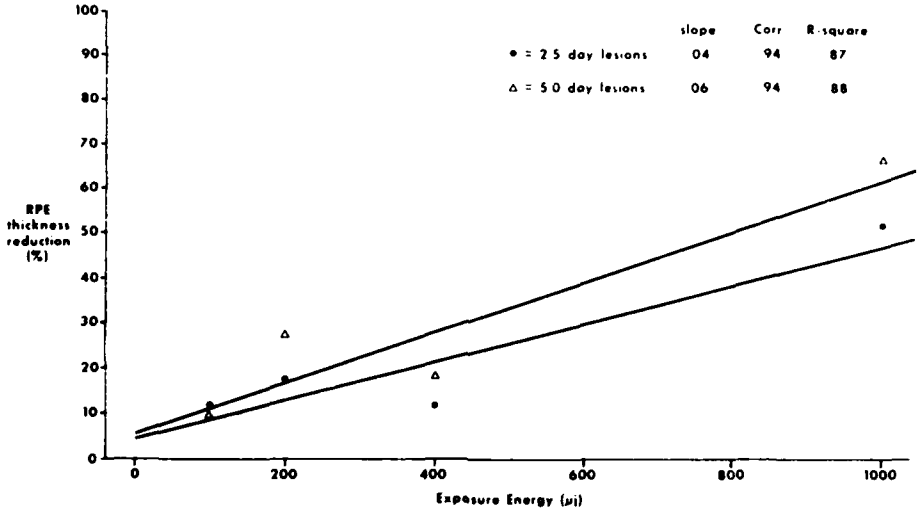

Figure 3. The effect of Q-switched ruby laser radiation on retinal pigment epithelium thickness in lesion centers.

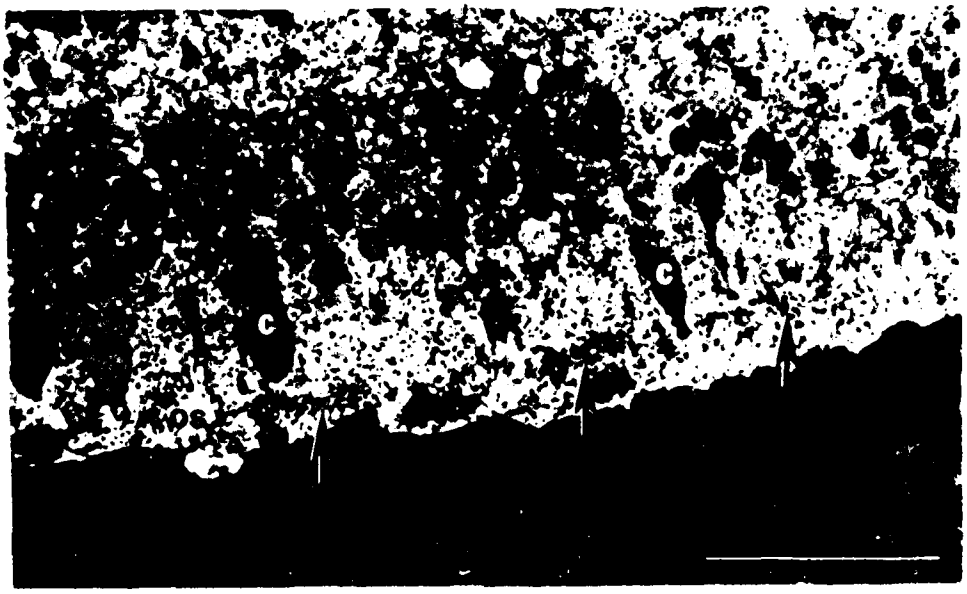

Figure 4. 1000 μJ, 2.5-day lesion. Lesion center is to the right. The following retinal layers are indicated in the photograph: outer plexiform layer (**op**), outer nuclear layer (**on**), photoreceptor inner segments (**is**), and outer segments (**os**). Cones (**c**) show increasing severity of inner segment degeneration and decreasing amount of label uptake left to right. Arrows indicate band of rod outer segment label which disappears on the right. The amount of label in the outer nuclear layer on the right is less than on the left. Hemorrhage (**H**). Bar = 50 μm.

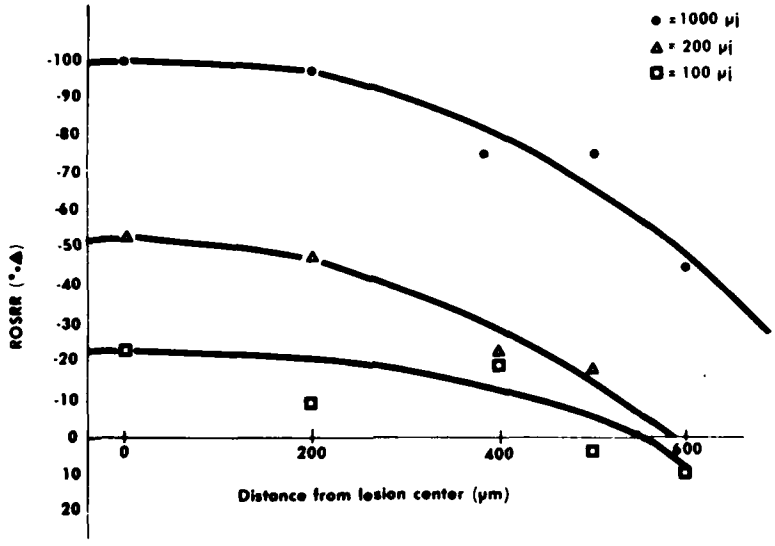

Figure 5. 2.5-day lesions show a decreased inhibitory effect with decreased exposure energy.

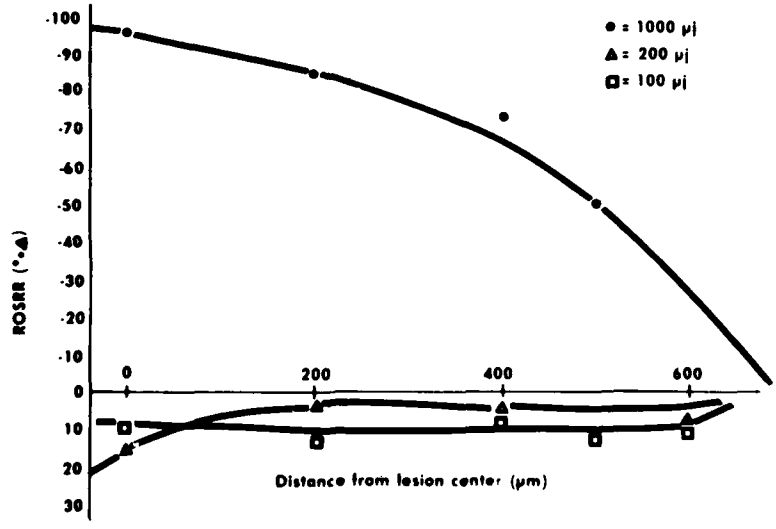

Figure 6. 5-day lesions show a recovery effect at the 200 and 100 μJ levels.

Table 2A

Location of labeled discs and length of rod outer segments

Exposure Energy (μJ)	Fundus Site	Lesion* (days after exposure)	Labeled disc† location (μm)	Outer segment‡ length (μm)
1000	Temporal	A 2.5	2.0	26.0
		B 5.0	2.3	30.0
400	Superior	A 2.5	2.3	30.0
		B 5.0	8.6	34.1
200	Nasal	A 2.5	5.6	30.0
		B 5.0	8.6	30.6
100	Inferior	A 2.5	7.1	35.0
		B 5.0	9.2	32.6

* 1 animal, 1 eye

† Distance from the base of the outer segment to the location of the center of the heavily labeled band (see Figure 2) in the center of the lesion (values represent the average of the center and 200 μm from the center data points).

‡ See Figure 2

Table 2B

Renewal time after injection of ^{3}H-leucine in Fundus

Fundus Site	Lesion* (days after exposure)	Rod outer segment renewal time (days)	
		Estimated time with exposure†	Estimated time without exposure‡
		lesion @ 1000 μJ	
Temporal	A 2.5	32.5	8.0
	B 5.0	32.6	9.3
		lesion @ 400 μJ	
Superior	A 2.5	11.4	9.3
	B 5.0	9.9	10.5
		lesion @ 200 μJ	
Nasal	A 2.5	13.4	9.3
	B 5.0	8.9	9.4
		lesion @ 100 μJ	
Inferior	A 2.5	12.3	10.8
	B 5.0	8.7	10.1

* 1 animal, 1 eye.

† Formula: (Outer segment length divided by distance from outer segment to center of labeled band) x (days after injection) = renewal rate. Measurements can be obtained from Table 2A

‡ Distance from outer segment to labeled band, based on findings in control = 8.1 μm. Same formula† is applied with 8.1 μm as divisor.

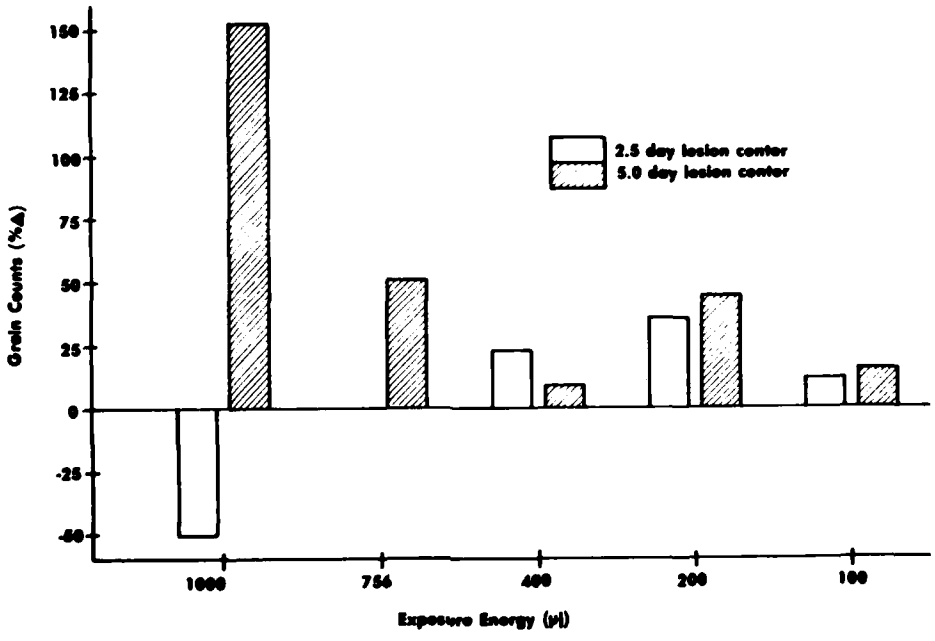

Figure 7. Grain counts over outer nuclear layer nuclei in lesion center as compared with the lesion periphery (600 μm from center).

reduction (-50%), while all the other lesions showed an increase. At 5 days, the ONLGCs showed an exposure-energy dependent trend, although not linear (counts decreased with decreasing exposure energy, i.e. 1000 > 200 > 100 μJ); however, these counts were elevated above normal more so than the 2.5-day counts. The greatest percent increase in the ONLGCs occurred in the 5-day 1000 μJ lesion (+153%); the macula showed the next highest increase (+51%).

A comparison of the percent increase in ROSRRs with the percent increase in ONLGCs indicates that all of the 5-day lesions had a back log of leucine in the outer nuclear layer (ONL).

DISCUSSION

Because disc formation is steady, it is possible to predict, from a single radiolabel injection, the length of time necessary for the labeled discs to be displaced along the entire length of a rod outer segment, and thus the time required for complete outer segment renewal. Two autoradiographic variables, ROSRRs and ONLGCs, gave remarkable insight to the extent of alteration as a function of laser exposure energy and as a function of the amount of recovery with time.

Tso et al (12) have shown that lasers preferentially alter cone morphology. In our study, we confirm that cones are damaged more

severely than adjacent rods. In addition, by comparing the relative amounts of inner segment label between adjacent rods and cones, cones showed less label uptake. Unfortunately, cone outer segment synthesis processes could not be evaluated, as membrane precursor proteins yield only a diffuse scattering of newly synthesized molecules throughout the existing membranes and photoreceptor (13). Nevertheless, our study appears to be the first to correlate both morphologic and metabolic sensitivity for cones over rods. As part of a recovery response some macular cones showed newly formed outer segment membranes in their inner segments. These have previously not been described for laser exposed retinas.

In the 5-day 200 μJ (ED_{50}) and 100 μJ lesions the upper limit of 10-15% above normal in ROSRRs indicated a maximum recovery response. Upper limits to membrane rod outer segment addition have been reported in other autoradiographic studies (14) to result from exposure to elevated ambient temperatures and from exposure to constant light. Several things are suggested by an upper limit increase in rod outer segment synthesis: 1) that a critical inner segment factor regulates outer segment synthesis and that this factor is rate-limiting; 2) that the upper limit reflects an increased aging process; and 3) that the upper limit in membrane synthesis may not be sufficient to meet the repair demands of the photoreceptors and as a result photoreceptor cytoplasmic efficiency would be decreased. This may manifest itself as the continued production of altered outer segment membranes many months after laser light exposure (2). Longer term studies need to clarify if this increased synthesis rate persists, increases, or returns to normal.

Increases in outer nuclear layer label are greater with increasing time after laser insult and with increasing exposure energy. Such data have not been previously reported and indicate that protein synthesis sites in the inner segment have been altered and that protein precursors accumulate in the inner portion of the photoreceptor. The more extensive the inactivation of membrane synthesis centers, as with increasing exposure energy in the inner segment, the greater is the amount of label shunting to the inner photoreceptor regions. Alternatively, the increased outer nuclear layer label after exposure may reflect increased synthesis due to repair processes. The large decrease below normal in outer nuclear layer label concentration 2.5 days after 1000 μJ exposure may represent a reduced efficiency in membrane transport function in addition to decreased protein synthesis. This is most likely related to the thermal effect of laser radiation.

Our results suggest that inhibition involves two primary photoreceptor processes: 1) the active transport of amino acids, or perhaps passive diffusion across membranes, and 2) inactivation of intracellular protein synthesis centers (i.e., ribosomes or critical enzymes). Floyd et al (15) have reported that Q-switched ruby laser

pulses ablated particles from the surfaces of membranes. Marshall (1) in an in vitro autoradiography study reported that inhibition of protein synthesis after Q-switched ruby laser exposure was thermally dependent and most likely occurred at the ribosomal level. It would seem reasonable then that inhibition of protein synthesis in membrane renewal may also involve the ablation of ribosomes from the rough endoplasmic reticulum and thus uncouples protein synthesis. Furthermore, if Q-switched ruby laser pulses ablate surface particles from membranes then alterations in the composition of membrane proteins, and therefore transport functions, could account for the reduced amount of label seen at 2.5 days after exposure in both the photoreceptor nuclei and outer segments. Alternatively, the concentration or activity of an enzyme(s) may be the sensitive endpoint.

The extent of inhibition and recovery as revealed by the shifting pattern of black grains, which represent membrane precursor molecules in retinal laser lesions, show that rod outer segment renewal rates are more sensitive to the 694.3 nm laser radiation than label uptake by nuclei. The large quantities of label accumulation in the outer nuclear layer may serve as a reservoir for the critical rate-limiting reaction in the inner segment. The maximum upper limit to synthesis in rod outer segments may continue for an unknown period of time in an attempt to deplete this reservoir which may be directly related to the repair demand. Whether or not this is the case will require further studies.

In general, inhibition of membrane renewal processes in this study is consistent with the thermal and mechanical damage theory of retinal tissue in laser lesions (1,16).

In summary, the morphologic findings are consistent with both the thermal and mechanical disruption of retinal tissue in laser lesions. Cones are both morphologically and metabolically more sensitive than rods to 694.3 nm radiation. In a recovery response, cones synthesize outer segments in their inner segments. An attenuation effect to the RPE and an increased absorption effect are suggested in the macula. Rod outer segment renewal rates are initially more sensitive to Q-switched ruby laser radiation at 694.3 nm than label uptake by the outer nuclear layer. Near- threshold lesions show a recovery response in ROSRRs by indicating a small maximum upper limit increase, while a large backlog of protein precursors accumulate in the outer nuclear layer. Inhibition in protein synthesis may occur at the ribosomal level, while recovery involves some critical rate-limiting step. Low-level (subvisible ophthalmoscopically) retinal laser lesions show an increased aging process and altered metabolism, which suggest an increased sensitivity of analysis with autoradiography and reevaluation of the ophthalmoscopically determined ED_{50}. The Army may discover that these findings may have implictions for retinal conditions detected in users of laser devices.

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